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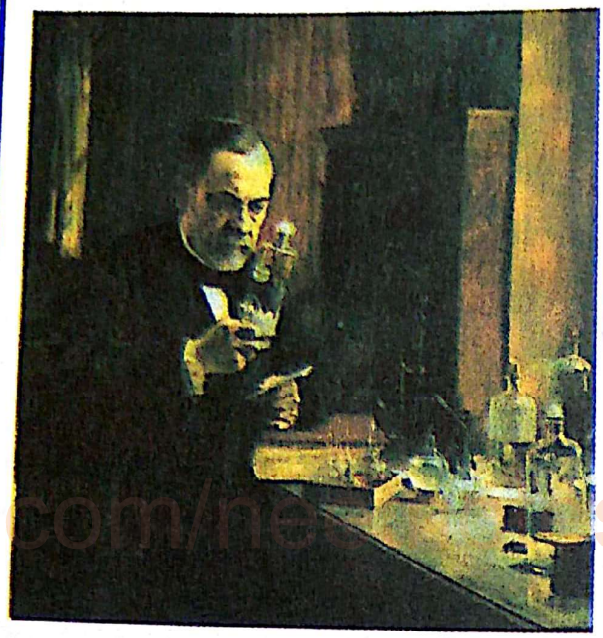
UNIT ONE

The Living World

If you look around in a forest or in an open field, or even along a roadside in a backyard, you will notice that the world abounds with astonishingly diverse living things. The living world exhibits a marvellous spectrum of colours, shapes and forms. You might recall your previous knowledge about many diverse forms of life. Life occurs almost everywhere on Earth, from the driest desert to the icy polar waters and from the highest mountain peak to the deepest ocean trenches. Some of these are microscopic and single-celled, while the others are many-celled. Biologists have identified and named a large number of species. You can imagine how rich a tapestry the living world is. Biologists focus on how life works. The living organisms possess distinct characteristics but display organisational and functional unity, entail a mechanism of origin and evolution of diversities and maintain a balancing relation with nature. All these aspects of life fall within the arena of the science of Biology. Jean Baptiste Lamarck first suggested the term biologie (biology) as the name for the 'new science' that he saw emerging from the fusion of the classical subjects of zoology and botany. In this Unit, you will learn about the nature of biology as science, gradual evolution of biology through ages, the various branches and scope of biology, importance of biology to overcome ignorance and resist its misuse and the major career options in biology. It will also unravel the basic characteristics and organisations of living world, including molecular organisation. This Unit will also enable you to understand origin of life and evolution of its diversity.



Louis Pasteur

(1822-1895)

Louis Pasteur, French microbiologist and chemist, studied in Paris. In 1867, a laboratory was established for him with public funds and from 1888 till his death he headed the Pasteur Institute. Pasteur proved that yeasts, which turn grape sugars into wine, are microscopically small living beings, and that fermentation does not require oxygen. This showed that basic life processes are chemical reactions. Pasteurization, to make dairy products free from germs, is based on his discoveries. Pasteur disproved the popular belief that microorganisms and other lower forms of life generate spontaneously from dust, rotting meat or dung. He proved that each microbe comes from an existing microbe. Pasteur was the first to establish the basic cycle of life that all living beings eventually become food for microorganisms, which, in turn, become fodder for new life. Pasteur concluded that microbes or germs cause disease (germ theory of disease). Pasteur saved the French silkworm industry by identifying two microbial diseases that were decimating the worms. He inspired his pupil Joseph Lister to work in antiseptic surgery. He also developed vaccines against rabies and anthrax.

Chapter 1

NATURE AND SCOPE OF BIOLOGY

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The study of living world is a special endeavour of humans. We humans are biological beings and are little different from other living organisms. Although several animals have curiosity, but its interpretation and verbal communication that makes humans singularly different from other living organisms. We keep pets, nurture plants, visit zoological and botanical gardens, climb mountain peaks or dive deep into the sea and undertake trekking in the forests to enjoy the serenity of wilderness along with its vast living and non-living resources. All these activities reflect the innate attraction of humans for natural world and their feeling of connectedness to living things. In fact, the scientific extension of human curiosity has given birth to the science of biology.

1.1 BIOLOGY : THE SCIENCE OF LIFE

The living organisms interact with one another as well as with their physical and chemical environment. **Biology** (*Bios*: life; *logos:* to discourse) is the science that studies living organisms — what they are? how do they work, interact and evolve? However, biological systems under the influence of nature show many exceptions. Scientists have to accept the biological exceptions with open mind because today's exceptions may not remain so in the light of tomorrow's knowledge and information. Biology as a discipline of science demands a scientific approach for analysis of its facts. It is, therefore, imperative that students of biology should have a clear idea about the basic nature of science.

1.2 NATURE AND METHODS OF SCIENCE

The word 'science' is derived from a Latin word *scientia*, meaning, 'to know'. Science is a process that is used to answer the questions about nature. A scientist links patterns or draws relationship among a number of isolated facts, determines principles from the observations on specific cases and, finally, discovers general principles. For example, Charles Robert Darwin (1809-1882) first made observations on variation of plants and animals. Next, combining his observations with his own experience in breeding domestic animals Darwin made some generalisations and set forth the ***Theory of Natural Selection***. This theory holds that plants and animals and their kinds change over time and is backed by a wealth of evidence. Evolution by natural selection is a particularly important theory since it provides the conceptual framework that unifies biology as a science.

A scientific bend of mind calls for the understanding of the basic nature of science— its method of approach to arrive at an inference. Suppose, your flashlight is not working. What has gone wrong ? May be the batteries are dead or the bulb is burnt out or the switch is out of order. In the extreme case, all of the three possibilities may be true at a time. What will you do? How will you proceed to solve the problem? You may try to replace the old batteries with new ones, or old bulb with a new one, or repair the switch. Thus, your action will follow a sequence of steps depending upon your preference. Firstly, you will formulate a provisional explanation or a **hypothesis** based on some rational variables, such as, the problem may be either with the batteries or with the bulb or with the switch. Secondly, in order to identify the actual problem you will test the hypothesis conceived by you for non-functioning of the flashlight. Ultimately, you will draw an **inference** about the actual problem with your light and get the fault corrected. Such a methodical inquiry is the essence of the scientific approach.

Generally, the scientific method of gathering information involves some steps. These are

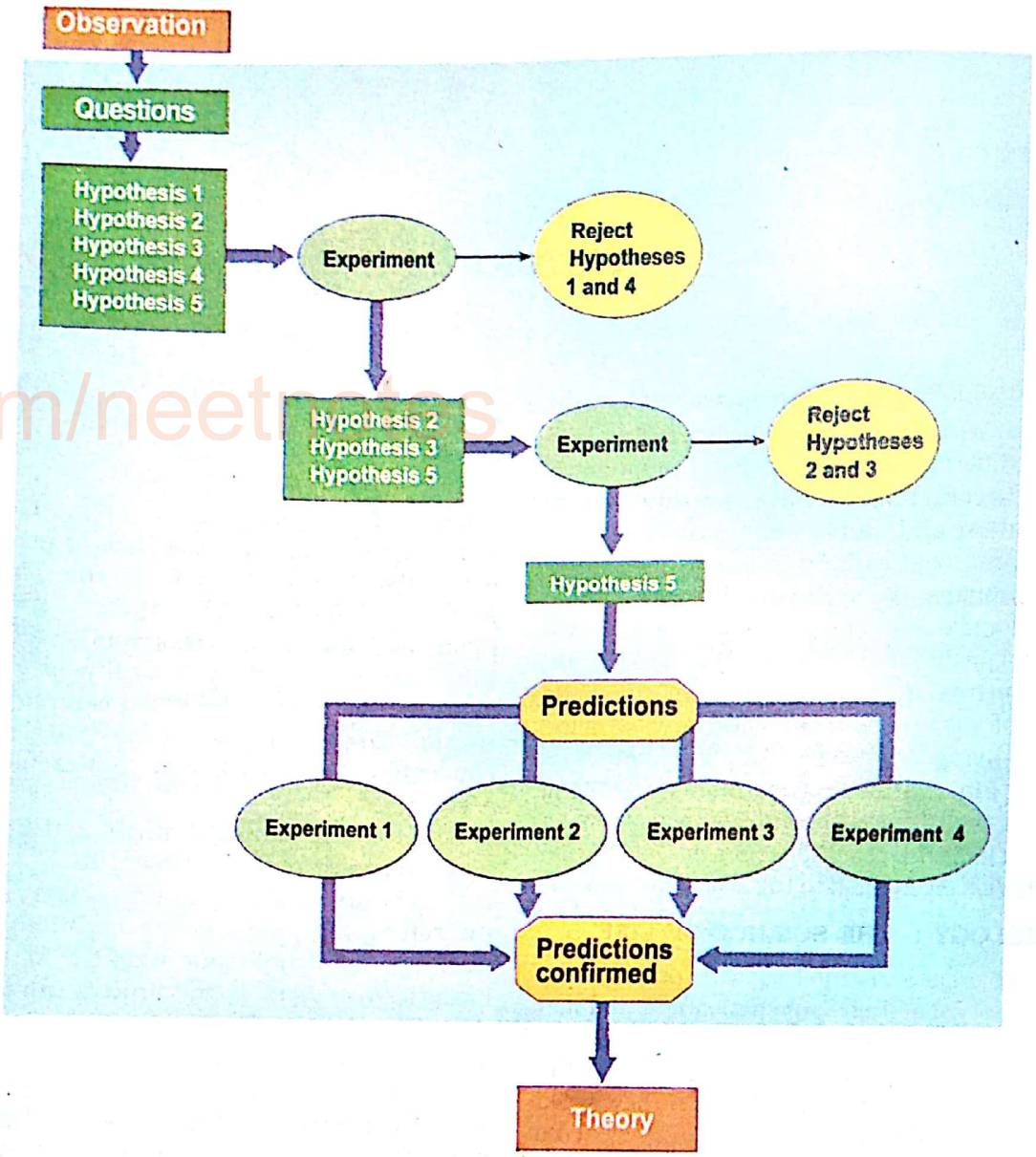

Fig. 1.1 *Generalised approach in science*

Observation, Formulation of Hypothesis, Testing of Hypothesis and Developing Theory (Fig. 1.1). Let us now take some examples from biology to understand the steps.

Observation

It is simply the ability to notice something, keeping records and finding out answer for questions framed by the observer. We can observe with our normal senses, even without taking the help of any instrument. For example, we can see, feel, smell, taste and touch things and discern the living world. Often we need microscopes, chemical analysers, or radiation detectors in order to extend our senses. Handling of a microscope needs considerable skill. Field binocular and tele-lenses are some of the other aids that may help us to observe distant plants, animals, birds etc. In this way, much information may be gathered for scientific studies. Just a magnifying glass can help you to observe and answer the question whether there is any living thing in a drop of pond water.

A scientific investigator must have a clear idea about what is to be observed and what is to be ignored. A scientist needs to direct the observations that raise questions, like 'what', 'why', and 'how'. Measuring everything is neither wise nor possible. It is also very important to find out the proper method of measurement and keeping

record of data. All data should be preserved because any one of them might be useful in answering any present or future question.

Formulation of Hypothesis

The next requirement is to develop a number of potential explanations or hypotheses based on the observation. Suppose, suddenly you realise that a room is dark. As an explanation for this perception you may formulate several hypotheses, such as the switches are turned off or the bulbs are burnt out or, perhaps, you are going blind. Actually, a hypothesis is a logical explanation that accounts for the observation. This step involves guessing or predicting the plausible answers to a question. A good hypothesis should be as simple as possible. A hypothetical proposition might stand true or false. Hence, the data that emerge from a set of observations must be analysed, which may help to make some kind of coherent generalisation.

Testing of Hypothesis

You need to test the hypothesis. For this, you must devise ways of testing. Generally, the scientists carry out an experiment for testing one or more hypotheses. Based on the result of experiment they eliminate or confirm one or more of the hypotheses. It is necessary to formulate **predictions** based on the remaining ones. Further experiments, if necessary, are to be carried out to test these predictions. This process helps to reject the most unlikely hypothesis and select the most likely one. Considering the hypotheses made earlier for explaining the cause of darkness in the room, first, as an experiment, you may try to switch on the lights. If the room is still dark then the first hypothesis is untrue and hence to be rejected. The shortcoming of this experiment is that it does not prove the other two hypotheses as true or false.

In another example, suppose, a scientist observes that the *Ginkgo* trees shed their leaves all at a time, around the month of November, and tries to find out a scientific explanation of this observation. The questions that might come to the mind of the observer – Why it is so that the leaves are shed all at a time ? What is the possible explanation of this observation ? One or more of the following predictions may appear to be the correct explanation:

(i) The plant may possess an internal rhythm or clock for timing its shedding of leaves.

(ii) The individual leaves are able to sense the day-length and each leaf responds independently and falls when the days are short enough with the onset of winter.

(iii) The fall of leaves might have been caused by a strong wind.

To eliminate one or more hypotheses the scientist might cover some leaves to prevent the use of light for sensing the day-length by the leaves. If these covered leaves do not fall but the uncovered ones are seen to shed, the second hypothesis becomes true. Alternatively, if the covered leaves are still observed to fall, then the second hypothesis becomes untrue and hence it is rejected. Next, if the fall of the leaves is not recorded to be associated with a gale, then the third hypothesis is also eliminated. Finally, the first hypothesis stands as the possible cause and provides explanation of simultaneous falling of *Ginkgo* tree leaves. As this experiment helps the scientist to reject the majority of the alternative hypotheses or predictions, this is a successful experiment.

A standard type of experiment is called **control experiment**. Usually, a scientist performs two sets of parallel experiments that are identical in all respects except in one variable. Consider the experiment of Louis Pasteur (1862) on spontaneous generation of life (Fig. 1.2). He took a few straight and long-necked identical flasks and then placed similar nutrient solution (broth) in them. After that he bent the necks of a set of flasks to make them swan-necked. Next, he boiled the broths in all the flasks of both sets in order to kill any micro-organism that might be present in the nutrient solution. Then, he allowed the sterile broths to settle for several weeks. In this experiment, the straight and long-necked set of flasks served as control.

Later on Pasteur found that bacteria and mould appeared in the straight flasks but not in the swan-necked flasks. Microbes entering the straight-necked flasks could contaminate the nutrients. The swan-necks acted as filters – any microbe-laden dust particles moving into the swan-necked flasks got trapped in the films of

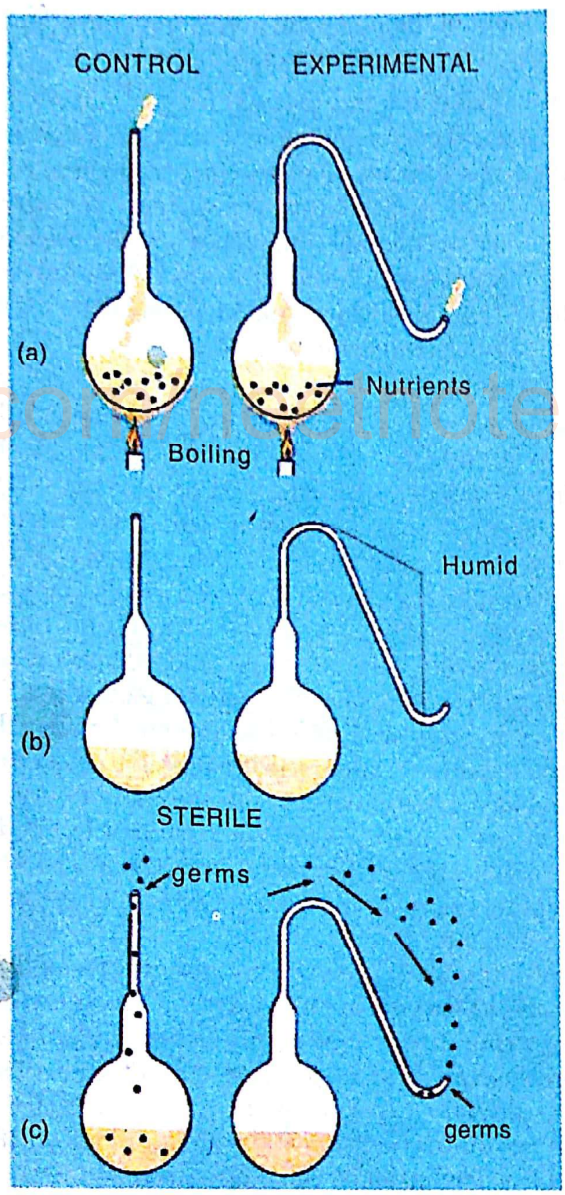

Fig. 1.2 *Experiment of Louis Pasteur (a) A long and swan-necked flask with infected broths (b) Broth in both flasks boiled to make it sterile (c) Reinfection of broth of long-necked flask, the swan-necked flask remains uninfected*

moisture accumulated on the humid curves of the necks. This simple experiment led Pasteur to counter the prevailing concept of spontaneous origin of life and finally put it to rest.

Following the same method you can perform many experiments. For example, to test the specific type of nutrient that affects the growth of plant, you may take five pots, each containing the same type of plant. Then add different nutrients to four of them and no nutrient to the fifth pot. This pot will serve as the control for your experiments. In the next step, expose all the five plants to sunlight and water them uniformly. Observe the differences in growth of all the plants, record your data and formulate the appropriate hypothesis.

Thus, science is a body of knowledge derived from observations and experiments, which are directed to determine the principles underlying what is being studied. It is a systematic and a precise objective way to study the natural world. It is also a process of minds that are creative, imaginative and social. When done correctly, science is neutral and unbiased.

Developing Theory

If the repeat experiments to test a hypothesis or prediction give the same result again and again the hypothesis gains validity. A hypothesis that survives repeated tests over a long period of time and that has central importance to an area of science may be considered a **theory**. Theories often comprise broad statements that cover large bodies of scientific knowledge. However, every experiment may not lead to a theory.

Formulation of a theory may require a long time. For instance, Robert Hooke observed the presence of **cellulae** (in Latin, small rooms and hence cells) as early as 1665, whereas the actual living cells were observed a few years later (1670) by Anton van Leeuwenhoek. Actually, Hooke's concept of cell received the proper recognition in 1838, when a botanist, Matthias Schleiden, carefully studied plant tissues and developed the first statement on ***Cell Theory***. Following this, Theodor Schwann reported animal tissues in 1839. An important extension of Cell Theory that all living cells arise from pre-existing cells. It came 20 years later in 1862 when Louis Pasteur successfully refuted the concept of spontaneous origin of life. Today, the likelihood of the cell theory being rejected is almost zero.

The above discussion reveals that the earlier knowledge acts as the foundation for a new finding. Therefore, any scientific finding should be published. Publication not only helps in the spread of knowledge to the world communities but also facilitates further investigation and enrichment of treasures of knowledge.

Science and Technology

In general, scientific investigations may be basic or applied. Basic science is intended to extend

the boundaries of what is already known. Thus, some scientists may find interest in discovering new plants or animals or studying comparative morphology or anatomy of living organisms. Applied research involves the application of scientific knowledge and technology for the benefit of humanity. These include the discovery of new drugs, production of improved varieties of stocks of economically important plants and animals, application of biotechnology for treating many human diseases and combating the poverty and food crisis, etc.

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Invention of new technology extends our ability to observe and measure. Technology enables scientists to work on questions that were previously unapproachable. For example, the knowledge of electromagnetic theory of physics has been instrumental in designing the electron microscopes and this invention has opened up the scope for gathering the present day knowledge about the structure of cells. Similarly, biotechnology, involving transport of foreign genes into micro-organisms, is the fruit of the discovery of molecular structure of DNA in 1953 by Watson and Crick, which has its basis on the knowledge of physics and chemistry.

Thus, the prevailing discoveries of science can be applied to invent new technology, which, in turn, may facilitate the advancement of scientific knowledge. In other words, information gathered from basic researches enriches the treasures of scientific knowledge and expands the field of applied researches. Also the fruits of applied research contribute to widen the field of basic researches.

1.3 BIOLOGY IN ANCIENT INDIA

The earliest human activity in the Indian sub-continent can be traced back to the early, middle and late Stone Age (400,000-200,000 BC). Actually, a major shift in human evolution occurred between 12,000 and 10,000 years ago when humans learnt to cultivate plants and domesticate wild plants and animals and, thus, developed agriculture and animal husbandry. The plants included wheat, barley, lentils and peas. Cultivation of rice was most likely achieved in India at Mehergarh and Mahagarh about 6,000 years ago. Wild rice originated somewhere in the coast of Bay of Bengal (Andhra, Orissa and Bengal). Ancient Indians developed at least 100,000 varieties of rice. Of these, many strains were nutritious, fragrant, salt resistant and flood resistant. They also bred and raised animals like goat, sheep, pig, cattle dog, etc. All such activities of the ancient people, presumably, required some degree of scientific attitude.

The people of Vedic ages (2500 BC to 650 BC) had a superb tradition of making observations on plants and animals and recording the same. Our Vedic literature recorded about 740 plants and 250 animals. The first attempt of classification is observed in *Chandyogya Upanishad*, which classified animals into three categories — *Jivaja* (Viviparous), e.g. mammals, *Andaja* (Oviparous), e.g. birds, reptiles, insects and worms, and *Udbhija* (Vegetal origin), e.g. minute animals. Post-Vedic Indian literature, such as *Susruta Samhita* (600 BC) classified all 'substances' into *sthavara* (immobile), e.g. plants, *jangama* (mobile), e.g. animals. Plants were further subdivided into *Vanaspati* (fruit yielding, non-flowering plants), *Vriksha* (both fruit yielding and tall flowering plants), *Virudha* (shrubs and creepers), and *Osadhi* (plants that die with ripening of fruits). Susruta described in detail the parts of plants, such as *Ankura* (sprout), *Mula* (root), *Kanda* (bulb or stem), *Patra* (leaf), *Puspa* (flower), *Phala* (fruit), etc. *Susruta Samhita* also mentioned about classification of animals, such as *Kulacara* (those herbivores who frequent the river banks, e.g. elephant, buffalo, etc.), *Matsya* (fish), *Janghala* (wild herbivorous quadrupeds, e.g. deer), *Guhasaya* (carnivorous quadrupeds like tiger, lion, etc.). *Susruta Samhita* also records some observations on snakes (both venomous and non-venomous) and leeches.

Ancient India made surprising advances in **medical science**. In fact, Indian medical tradition goes back to Vedic times when the two Ashwinikumars practised medicine. At that time Dhanvantari was regarded as the God of Medicine. Actually, Susruta is one of the earliest scientists who studied human anatomy. He has described, in detail, anatomy based on his studies on dead body. *Susruta Samhita* is considered as the oldest treatise on surgery. Susruta himself carried out plastic surgery of human nose **(rhinoplasty)**. He used non-poisonous live leeches for preventing clotting of blood in the post-operative cases. Now we know that leeches actually secrete **heparin** along with saliva for such purpose during feeding of blood. Susruta also specialised in **ophthalmic**

surgery (extraction of cataracts). He is, therefore, acknowledged as the 'Father of Surgery'. The other ancient Indian medical practitioners included Atreya (600 BC) and Caraka (100 BC). *Caraka Samhita* (100 BC) is truly a revised edition of an encyclopaedic treatise originally written by an earlier physician, Agnivesa under the guidance of Atreya. Caraka was the first physician to present the concept of digestion, metabolism and immunity. According to him, a body functions because it contains three doshas, namely, bile, phlegm (spit) and wind, and illness is caused when the balance among the three doshas in a human body is disturbed. Caraka prescribed medicinal drugs for restoring the balance. Caraka knew the fundamentals of genetics. For instance, he knew the factors determining the sex of a child. The indigenous system of medicine in India is known as **Ayurveda** (*Ayu* means life; *veda* means knowledge) which is the science of living or longevity. It developed by taking ideas largely from the doctrines of *Caraka Samhita*. Ancient Indians also succeeded in understanding the origin and evolution of life in its broad outline. Caraka held that the individual is a replica of the universal spirit. Both man and the visible world are composed of six elements: *Prithvi* (earth), *ap* (water or liquid), *tejas* (fire), *vayu* (air) and *akasa* (ether). The sixth element, the spirit or self in the individual, is equivalent to Brahman in the universe. *Ayurveda* deals with the origin of life. The *Taittiriya Upanishad* (ca 7-8 BC) has made important observations on the evolution of life. It traced the evolution of life to space. Besides, Manu's texts in Sanskrit, *Manu Samhita* or *Manu-smriti* (200 AD) propounded evolution.

1.4 ARISTOTLE : AN EXCEPTIONALLY TALENTED PERSON

Aristotle (384 - 322 BC), a great philosopher of Greece, belongs to time when biology as a science was poorly developed. He was a very careful and meticulous scientist as well. He spent years studying the natural sciences and collecting specimens. Aristotle relied on observation and contributed much in biology. About 90 per cent of his writings are on scientific subjects, mostly on biological ones. A few important contributions of Aristotle are noted below:

(i) Classified animal species and arranged them into hierarchies. His mode of classification was reasonable and, in some cases, strikingly modern.

(ii) Formulated the concept of the **Great Chain of Being** or ***Scala Naturae*** — a chain of progressive change in nature. This corresponds to a sort of evolution.

(iii) Dealt with over five hundred types of animals and dissected nearly fifty of them.

(iv) Studied the developing embryo of the chick.

(v) He reported that sharks give birth to live young (viviparous) but do not develop a placenta like mammals.

(vi) Observed the placenta in dolphins as a means of nourishment to the foetus. Based on this similarity with mammals he classed dolphins with mammals.

1.5 EMERGENCE OF CONTEMPORARY BIOLOGY

Despite the significant contributions of Aristotle, biology did not develop as a scientific discipline before the early sixteenth century. Nevertheless, since then the works of many scientists have gradually laid the strong foundation for the emergence of contemporary biology.

The earliest record of scientific approach in biology is that of a Belgian scientist, Andreas Vesalius (1514 -1564). His treatise is named *De Humani Corporis Fabrica* (The structure of human body). In this book, he has mentioned that human body is composed of many complex 'subsystems', each with its own function. Andreas Vesalius is honoured as the 'Father of Anatomy'. It was William Harvey (1578 - 1657), a British scientist, who first demonstrated that the heart pumps blood and that the blood circulates. His monograph was named *Anatomical Exercise on the Motion of the Heart and Blood*. He also contributed on the reproduction and embryonic development of chick.

The third of the pioneers was Robert Hooke (1635-1703), a British scientist, who first coined the term cellulae in 1665 and this term is synonymous to what we call cells. His book was named *Micrographia*. Actually, Hooke observed a thin slice of cork under his self-made simple microscope. He recognised some honeycomb of empty, tiny 'microscopical pores' or compartments. A few years later, in 1670, a Dutch cloth merchant turned into a scientist, Antony van Leeuwenhoek (1632 -1723), observed the real living cells with the aid of a 'simple microscope'

made by him. He is regarded as the inventor of 'simple microscope'. He was the first to draw a diagram of bacteria in 1683. Besides, he contributed a lot about different types of cells, such as *Euglena*, ciliates and cells like sperm, eggs and blood corpuscles of invertebrates. Also he reported about compound eyes of insects and many other animal structures.

Aristotle's work on classification went unchallenged until 1753 when Carolus Linnaeus (1707-1778), a Swedish naturalist, published a book *Species Plantarum*. This book registers about 6,000 species of plants. Also he published a book *Systema Naturae* in 1758. This book recorded more than 4,000 species of animals. Linnaeus introduced the method of naming of plants and animals known as the **Binomial Nomenclature**. According to this method, the scientific name of a species consists of two parts — the first part represents the genus to which the species belongs while the second part represents the identity of the species to which the individual belongs. For example, humans are named *Homo sapiens* and the pea plants are *Pisum sativum*.

Until the beginning of nineteenth century, biologists had no idea about the changeable nature of species. Aristotle's *Scala Naturae* was the only to explain the cause of diversities of the living things. In its essence, a rat was born as a rat and a frog as a frog, and so on; all creatures remained unchanged since they were created as per Wisdom of God. It was Georges Leopold Cuvier (1769-1832), a French palaeontologist, who first rejected the traditional *Scala Naturae* as a unifying concept of evolution. Also, he was the first to identify the fossils of extinct bird-like reptile and laid the foundation of **palaeontology**, the study of fossils, as a branch of biology. Cuvier also made major contributions in **comparative anatomy**.

Jean Baptiste Lamarck (1744-1829), a French naturalist, was the first to discard the idea of 'fixity of species'. His book *Philosophie Zoologique* was published in 1809. Yet he could not offer sufficiently convincing explanation in favour of evolution. Although Leeuwenhoek observed the real living cells in mid 1670s, the first statement on Cell Theory came out much later in 1838 from Matthias Schleiden (1804-1881), a German botanist. He proposed the theory on the basis of his studies on plant tissues. Theodor Schwann (1810-1882), a German zoologist, further strengthened the theory in 1839 by his report on animal tissues. Charles Robert Darwin (1809 - 1882), a British naturalist, is the pioneer in the field of biology in the nineteenth century. His monumental treatise *On the Origin of Species by Means of Natural Selection : The Preservation of Favoured Races in the Struggle for Life* was published in 1859. Darwin's conclusion was sharply different from conventional wisdom. He elaborated the concept of evolutionary change and established that **Natural Selection** is the agent that brings about changes to result in the **Origin of Species** by evolution.

In 1862, Louis Pasteur (1822–1895), a French scientist, countered the prevailing concept of spontaneous origin of life. Also he proved that fermentation is caused by living organisms (yeast and bacteria) and firmly established the ***Germ Theory*** of diseases. He discovered the vaccine against anthrax caused by the bacterium *Bacillus anthracis* in cattle. His technique of killing of germs, a process of sterilisation, is known as **Pasteurization** and is widely used for sterilisation of bottled or pouched milk.

It was Gregor Johann Mendel (1822 -1884), an Austrian monk, who after eight years of careful and scientific cultivation of pea plants (*Pisum sativum*) discovered the principles of inheritance in 1865 and it was published in 1866 in a little known journal of the society. Although Mendel is regarded as the 'Father of Genetics', his extremely valuable theory remained unknown to the scientific world till 1900. As Darwin was quite unaware of Mendel's Laws of inheritance, he could not provide a satisfactory explanation of the mechanism of variation observed by himself in nature. In his book *On the Variation of Animals and Plants under Domestication* (1868), Darwin put forward his own ***Theory of Pangenesis*** as the mechanism of inheritance. According to this theory, every organ of the body produces minute hereditary particles called **pangenes** or **gemmules**. Darwin suggested that the gemmules were carried through the blood from every organ of the body and were collected together into the gametes. However, both Lamarck's concept of Inheritance of Acquired Characteristics and Darwin's theory of Pangenesis were rejected with the discovery of the ***Theory of Germplasm*** in 1892

by August Weismann (1834-1914), a German biologist. Weismann's experiment on rats established that the germ (sex) cells are set apart from the body (somatic) cells early in the embryonic development, and it is only the changes in the germplasm that can affect the characteristics of future generations.

In addition to the works of Darwin, Pasteur, Mendel and others, many revolutionary discoveries, made in the twentieth century, have led to the emergence of the contemporary biology. With its further development, biology has now emerged as the most demanding of all sciences. This is so partly because life is complex and radiant and partly because it incorporates the mechanics of physics, chemistry and engineering. The important breakthroughs made by scientists of twentieth century are summarised in Table 1.1.

1.6 WHAT DO BIOLOGISTS STUDY?

A biologist's concern is many-fold. Basically, the interests of biologists move around the coordinated functioning of living things. Biology is the science that deals with living things. Traditionally, biologists were used to concentrate either on the study of animal life or on the study of plant life. Consequently, biology has been divided into two major branches—Zoology, dealing with animal life, and Botany, dealing with plant life. The invention of the **microscope** in the sixteenth century gave a great momentum to biology by broadening and deepening its scope and incorporating the study of microscopic forms of life. This created the science of **microbiology.**

Table 1.1 Some Significant Contributions in Biology

Name of the Scientist	Year	Major Contributions
Hugo de Vries (1848–1935), a Dutch geneticist, Erich von Tschermak-Seysenegg (1871–1962), an Austrian geneticist and Karl Correns (1864 – 1933), a German geneticist	1900	rediscovered Mendel's contribution and this marks the beginning of the science of Genetics.
William Bateson (1861–1926), a British biologist	1909	conducted experiments that reinforced the idea that each trait is controlled by a different gene, discovered linkage and introduced the term Genetics.
Walter Sutton (1877–1916), American geneticist	1904	investigated chromosomes and discovered the Chromosomal basis of heredity.
Thomas Hunt Morgan (1866–1945), an American geneticist	1910–1930	described the phenomena of linkage and crossing over and created the first gene maps to show the linear arrangement of genes along the chromosome.
Alexander Fleming (1881–1955), a Scottish bacteriologist	1928	discovered penicillin, the toxic product of the blue mould *Penicillium notatum* that contaminated his culture of the bacterium, *Staphylococcus*. Penicillin is the first antibiotic drug and it was first used to cure soldiers in World War II.
Oswald Theodore Avery (1877–1955), American bacteriologist	1944	discovered that genes are composed of the nucleic acid called deoxyribonucleic acid (DNA). Since then DNA became the focus of genetics.
James D. Watson (1928–) an American biophysicist and Francis H. C. Crick (1916 – 2004), a British physicist	1953	discovered the double helical structure of the life, deoxyribonucleic acid (DNA). This discovery has triggered a new era of molecular biology.

Har Gobind Khorana (1922 –), born in Raipur, West Punjab, now in Pakistan, Robert W. Holley (1922 – 1993) and Marshall W. Nirenberg (1927 –), both from America	1968	shared the Nobel Prize in Physiology or Medicine "for their interpretation of the genetic code and its role in protein synthesis." Khorana is also the first to synthesise strings of nucleotides (the basic units of DNA), called oligonucleotides, in the laboratory. He produced the first man- made gene in his laboratory in the early seventies. These custom designed pieces of artificial genes are widely used in biology labs for sequencing, cloning and engineering new plants and animals. The concept of genetic code has equipped us with an in-depth understanding of the different aspects of biology at molecular level.
Norman Ernest Borlaug (1914 –), an American agronomist	1970	won Nobel Prize for his "green revolution", which involves the use of improved wheat seeds, new types of higher-yield rice and more efficient use of fertiliser and water. Modern Plant and animal breeders can now produce new forms of life with almost any mix of characteristics by altering the genes in DNA.
Stanley Cohen (1922 –) and Herbert Boyer (1936 –), both American biochemists	1973	discovered recombinant DNA (rDNA) technology and this marks the birth of modern biotechnology. In this method, researchers have been able to use genetically engineered bacteria to produce insulin, a hormone for treating diabetes. It is now possible to cure genetic diseases either by repairing the defective gene or introducing normal functioning genes. Scientists are now able to produce genetically modified (GM) crops and livestock.
Ian Wilmut (1944 –), a Scottish embryologist	1996	created the first boar calf clone from a frozen embryo, which he named Frosty. Later, in 1996, Wilmut and Keith Campbell produced the first live, healthy sheep clone, a Finn Dorset lamb, Dolly, from the differentiated adult mammary cells.
U.S. Department of Energy and National Institute of Health	2001	launched the Human Genome Project in 1990. It aimed to identify all the approximate 30,000 genes in human DNA and determine the sequences of 3 billion chemical base pairs that make up human DNA. All such information would be stored in databases for analysis. The draft of Human Genome has been published in February 2001.

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The scientific classification of organisms including their naming and identification fall within the realm of the broader science of **taxonomy**. Classification in biology is the systematic categorisation of organisms into a coherent scheme. Linnaeus in 1753, classified organisms according to their structural similarities, laying the foundation for modern scientific classification. The original purpose of biological classification or **systematics**, was to organise the vast number of known plants and animals into categories that could be named, remembered and discussed. Modern classification also attempts to show the evolutionary relationships among organisms and forms a part of taxonomy, which is the study of the relationships of organisms. It includes collection, preservation and study of specimens, and also analysis of data provided by various areas of biological research. There is an International Code for Classification and System of Naming of Organisms. Every taxonomist has to follow this method of classification. Assigning of names to organisms and to the categories in which they are classified is called **Nomenclature**.

Morphology is the aspect of biology that deals with the study of body structure of various organisms, including humans. Comparative anatomy is concerned with the structural differences of plant and animal forms. The study of similarities and differences in anatomical structures forms the basis for classification of both plants and animals. The study of morphology of internal systems or organs constitutes what is called **Anatomy** or **Internal Morphology**. An anatomist often finds that the internal structure is related with habits and habitats of organisms. For example, both human and cattle have stomach. But the cattle, being a herbivorous animal, possess a special part of stomach known as rumen, that suits their mode of feeding and digestion.

Offspring always resemble their parents. Why do you look like your mother or your father or to some extent both of them? What makes a person appear as he or she is? Why do the same diseases tend to occur in family members? Questions such as these could not be answered without the help of *Genetics*. *Webster's Concise English Dictionary* defines genetics as the science of heredity. This does not explain what genetics is actually concerned with and how much it has contributed to our knowledge about heredity, DNA and genes. DNA contained in the chromosomes is the determinant of heredity; it is transmitted to the offspring from the parents. The branch of biology that deals with the mechanism of inheritance and maintenance of hereditary characters of the organisms constitute the science called **Genetics**. Knowledge of genetics also helps us to account for the production of variation among organisms. Actually, genetics deals with the study of **genes**.

Organisms change in colour, form, and properties through time. Life evolves. Since its appearance in the primeval Earth, life has changed through evolution. Biology is a historical science that involves the study of living systems as they change through time. This theme of biology ties all living things together and brings forth the discipline called **Evolutionary Biology**. The history of life is a tale of the earth inhabited by changing cast of living things. The past history of living beings is preserved in the forms of fossils in the earth crust. In other words, fossils are the preserved remains, tracks, or traces of organisms that lived in the past. The fossils contained in layers of sedimentary rocks reveal a history of life on earth and provide good evidence in support of the concept of evolution. The study of fossils is called **Palaeontology**.

Another approach in biology is to study the relationships of organisms to their physical environment in which they live and to one another. This branch is called **Ecology**. The study of an individual organism or a single species is termed **autecology** whereas the study of groups of organisms is called **synecology**. A study restricted to ocean plants and animals and their ecological relationships is **Marine Biology**.

Biologists traditionally study the structural organisation of living things at tissue level. This branch of biology is called **Histology**. For routine histological studies biologists employ microtechnique, the first step of which is fixation of tissues. Live tissues are fixed in order to maintain their main architecture, that is protein. Therefore, fixation is often defined as stabilisation of protein. This involves treatment of the tissues with certain chemicals called fixatives. Fixation is followed by dehydration, paraffin embedding and slicing of tissues with an instrument called microtome and then staining of the cellular parts. At the end, the tissue sections are mounted and observed under a microscope.

The branch of biology that helps the biologists to understand the normal functioning of animals and plants during life and of the activities by which life is maintained and transmitted is referred to as **Physiology**. Normal functioning is based fundamentally on the activities of protoplasm. The study of function is usually undertaken along with a study of structure, the two being intimately related. Since the discovery of the cell structure, the science of physiology has undergone rapid development. It includes the study of vital activities in cells, tissues and organs. It also deals with processes such as contractility of muscle tissue, coordination through the nervous system, feeding, digestion, excretion, respiration, circulation, reproduction, and secretion. Virtually every specialised field in the biological sciences involves a consideration of the physiological aspects of its subject. **Plant physiology** includes also the study of photosynthesis and transpiration. As a separate and specialised branch, Plant physiology, arose

from attempts to apply the findings of **Animal physiology** to plants, and in its turn, contributed to the development of general physiology, especially in the study of cells. Studies on the structure and composition of cells are often referred to as **Cytology**. Studies attempting to correlate the structure of cells with their function, including the life processes of the cells, portray the branch of biology called **Cell Biology**. It is concerned with the organisation and functioning of the individual cell and depends greatly on biochemical techniques.

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Application of the knowledge of physics and use of the principles of resolution and magnification have resulted in the invention of compound light microscope. Here, light is refracted and directed to the eyepiece, where it is focused for the eye or a camera. It helps the biologists to gather certain levels of information about cells and tissues. But the finer details of the organisation of cells have only been possible to know with the aid of electron microscope. Electron microscope has its basis on the electromagnetic theory of physics. Having a higher resolution power it can magnify a cell up to 100,000 times. Such application of the knowledge of physics in the field of biology helps us to reveal the physics of life processes and, thus, emerged the branch of science called **Biophysics**. Biophysics is the application of various tools, techniques, methods and principles of physical science to the study of biological problems. These techniques facilitate the biologists to determine the structure of molecules in plants and animals to a degree not readily possible with ordinary chemical methods.

The physico-chemical composition of the inorganic and organic molecules of the cells and their interactions control the nature of cells. Studies related to the molecular organisation of organisms are regarded as **Molecular Biology**. It was W. T. Astbury (1898-1961), a British scientist, who first used and defined the term 'molecular biology' in 1950. Another interdisciplinary science called **Biochemistry** has emerged to consolidate the knowledge of organic chemistry and biology in a common place. Biochemistry is concerned chiefly with the chemistry of biological processes for elucidation of the living system. This branch of science has also been variously referred to as **Physiological Chemistry** and as **Biological Chemistry**.

Philosophical speculation that there might be other worlds similar to ours dates back to the ancient Chinese and Greeks. However, the achievements of space exploration technology and application of the physico-chemical principles for unravelling the molecular organisation of living being have motivated the scientists to test this speculation with experimentation. Many biologists are now working in collaboration with astronomers in search of life throughout the universe. This field of biology is called **Exobiology**.

The study of animal behaviour based on the systematic observation, recording, and analysis of how animals function, with special attention to physiological, ecological and evolutionary aspects is designated as **Ethology**.

1.7 SCOPE OF BIOLOGY

The current century is set to experience an explosion of information from the field of biology. People having the knowledge of modern biology are increasingly in demand and will continue to be so. Biology affects our daily lives and our future too. Many scientists are working on biological problems that critically affect our health and lives. Cancer, food production, poverty, population explosion, AIDS, global warming — these are the burning issues associated with human welfare. The full range of topics covered in the biological sciences includes some of the most important aspects that affect human life on earth as well as in space. The development and use of modern medicine depends on the understanding of cell structure and microbiology. Understanding of the functions of tissues, organs and organ-systems of the human body is also important in this respect.

Biologists study systematics and classification of organisms. This is very useful in modern medicine. It is important to know exactly which organism is causing a particular disease. A general notion of the type of organism is simply not good enough. Unless one knows the nature, life history and the name of the vector that spreads the disease, prevention and cure is not possible. For example, the female anopheles mosquito is a vector of the germ *Plasmodium*. This protozoan parasite causes the disease malaria in humans. By understanding the classification of

insects, the female anopheles mosquito can be identified. Knowledge of its life history may help to control and possibly eliminate the disease. Similarly, knowledge on the classification and life cycle of different species of parasitic animals, such as *Plasmodium*, *Entamoeba*, *Giardia*, *Ascaris*, etc., is of great value in combating human diseases.

Plants have medicinal importance. Medicines like Penicillin, Quinine, Kalmegh, *Nux vomica* are the natural products. Hence, a systematic knowledge of botany can help to discover many unknown plants of medical importance. Knowledge gathered on the different types of plants, especially their medical importance, is an advantage to a biologist for serving a human cause. A student of biology can also develop the necessary skill to understand the cause of many hereditary diseases and genetic disorders. With this basic strength he or she can offer services to human causes by way of genetic counselling. Also the knowledge of genetics can be used to help in the improvement of reproductive health of humans and combat the problem of population growth.

Courses in biology can equip the students with much more than understanding issues related to health. Organisms are diversified and at the same time are unified living beings. What is life? What are the features that make living things diverse? How do they exhibit unity? How do they perform the various functions of life? These kinds of questions can only be answered based on the knowledge of biology. Such courses as Anatomy and Physiology help people become aware of the structure and functions of the human body. A biologist can learn things related to economic uses of plants and animals. There are many plants and animals of economic importance. A student from biological stream can choose to specialise in Agriculture, Horticulture, Jute technology, Tea technology, Fishery, Aquaculture, Apiculture, Sericulture, Poultry farming, etc. These specialised abilities are useful to help in the development of national economy.

With modern biotechnology scientists have been able to produce **genetically modified (GM)** crops. It is believed that these crops will help the human society to do away with the problem of food shortage. Geneticists, evolutionists and ecologists, independently or collectively, can work to assess the efficacy of biotechnology and bring forth appropriate enlightenment to the modern society. The future directions of biotechnology, conservation of biodiversity, maintenance of environment and human welfare remain in the hands of biologists.

Biology helps students to understand how humans fit into the world of life as part of the earth's ecosystems. A student of biological discipline is wellequipped to understand the world of life. It is a modern tragedy in present day world that the majority of people are generally unaware of the value of other life forms and how these forms interact with one another. A biologist can well understand ecosystems and the balance of nature. The very survival of the human species depends on our ability to understand thoroughly the ecosystems of the earth. Evaluation of renewable resources, such as the biodiversity, is immensely important for human existence. A student of biology may cope up with wildlife and nature study with great ease. Courses such as Ecology, Marine Biology, Botany, Zoology, Environmental Science and Wildlife fulfil valuable goal of understanding nature.

1.8 BIOLOGY IN DISPELLING MYTHS, MISCONCEPTIONS AND DISBELIEFS

A large number of myths, misconceptions and disbeliefs exist about the living world. As a student of biology one can help general people and the society to overcome these menaces. A few examples are discussed below :

(i) A popular myth is that the snakes can hypnotise or charm their prey. The basis of this myth lies in the observation that rat, bird and other prey become frozen or motionless, perhaps, out of fear when confronted with a snake. Perhaps, the inability of snakes to blink the eyes has generated this myth.

(ii) It is also believed that a snake charmer can make the snakes dance on the tune of his flute. This is totally a misconception about the power of the snake charmer and the hearing apparatus of snakes as well. A student of biology knows that snakes have no external ears and so a poor hearing.

(iii) A common superstition, especially among the farmers, is that the snakes suck milk from the teats of cattle. Biology brings to light that a snake can drink by submerging its head

or mouth in water. It takes water by expanding its body wall. Above all, snakes do not have any mechanism for sucking. Being carnivorous they do not drink milk. Why the snakes move around barns? The answer is known to biologist. The rodents (rats, mouse, etc.) often inhabit the barns to eat rice or other grains and the snakes often visit that place in search of their prey, such as the rodents.

(iv) There is a common myth that carnivorous plants are "man-eaters". This myth has its basis in the biological fact that the carnivorous plants like *Dionaea* (Venus fly trap), *Nepenthes* (Pitcher plant), *Drosera* (Sundew), *Utricularia* (Bladderwort), etc., lure their live prey (insects) with odours and direct them for trapping inside their modified leaves. To be precise, some plants are carnivorous but not 'man-eaters'.

(v) There are various misconceptions and misbeliefs related to human illness. The very name of the disease malaria, meaning bad air, has its origin on the misconception that one can "catch" the disease when the air is "bad". This misconception or myth has been put to rest by Sir Ronald Ross, who worked at Calcutta (Kolkata) and discovered first, in 1897, the role of anopheles mosquito in the transmission of the malarial parasite. Biology tells us that malaria is caused by the protozoan parasite *Plasmodium*, which requires two hosts to complete its life cycle. The female anopheles mosquito acts as the vector of the parasite and infects vertebrates and humans with the germ. Have people in general accepted this biological basis of malaria? Why many of us disregard the advice of the health organisations for the control and prevention of malaria?

(vi) The most serious misconception and disbelief that rock the society is the notion about the **Acquired Immuno Deficiency Syndrome (AIDS)** of man caused by **Human Immunodeficiency Virus (HIV).** Immuno deficiency is the weakness of immune system, which normally fights against the infection. Syndrome means group of manifestations of diseases. A person suffering from AIDS faces not only social boycott but also public outrage. Some common myths about HIV infections are that the disease can spread even by sharing food with a person having AIDS. Why are the common people and some of those associated with medical practices are afraid of interacting with HIV positive persons? The answer is very simple. The former section is unaware of the cause of AIDS, not to speak of the HIV. The section of medical practitioners and nurses who refuse to treat or nurse the AIDS patient suffer from gross disbelief in science. Biology teaches us that HIV is transmitted only by direct contact with body fluids. Such contact may occur during transfusion of blood to a normal patient or sharing a common needle for drawing blood or injecting drug. Sexual contact may also result in direct contact of body fluids. Even an infected mother can pass the germ to her baby through placenta in the prenatal stage and breast milk in the postnatal stage.

1.9 MISUSE OF BIOLOGY

Biological Sciences have deciphered many techniques for the interest of human prosperity. But it is often seen that the biological techniques are wrongfully used by various agencies against the human race. Biologists can create awareness about the impact of misuse of biology. A few examples are discussed below :

(i) **Amniocentesis** is the removal of amniotic fluid via a needle inserted through the abdomen into the uterus and amniotic sac, in order to gain information about the foetus. The amniotic fluid contains cells (amniocytes) of the amniotic membrane and some foetal skin cells. These cells are then cultured and stimulated to grow. After a few days, the cells are broken to release the chromosomes, which are stained and counted and compared with the 23 pairs of normal human chromosomes for detecting missing or extra pieces. The centres for genetic counselling offer amniocentesis on request to women for such chromosome analysis. The amniocentesis technique has been developed for detecting foetal abnormalities by analysing **chromosomal defect** or **aberration** of the foetus. With the realisation that the test could reveal the sex of the foetus, people are seen to take the test mostly for knowing the sex of the foetus instead of the possible genetic anomalies. In many cases, the patients go

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or are forced to go for an abortion if the expected child is a female. This is a clear case of misuse of biological technique.

(ii) Alarmingly, biological techniques are being increasingly misused to produce lethal variety of infective agents for using them as **bioweapons**. Such misuse includes the development of antibiotic-resistant micro-organisms with increased infectivity. For example, anthrax is an acute infectious disease caused by the spore-forming bacterium *Bacillus anthracis*. Spores of *B. anthracis* can be produced and stored in a dry form keeping them viable for decades in storage or after release. A cloud of anthrax spores, if released at a strategic location to be inhaled by the individuals under attack may act as an agent of effective weapon of **bioterrorism**. An attack with bioweapons using antibiotic-resistant strains would, thus, initiate the incidence and spread of communicable diseases, such as anthrax and plague, on either an endemic or epidemic scale.

Biologists need to play an active role in creating awareness about the impact of misuse of biology on the human society and the living world.

1.10 CAREERS IN BIOLOGY

To make a good career in Biology, a student must study Biology along with Physics, Chemistry and Mathematics. This is the minimum requirement for taking higher education in the science general stream, such as Zoology, Botany, Physiology, Microbiology, Fishery, or Anthropology. After passing 10+2 course, students may choose any professional line and study Medical Science, Engineering or Technology.

Instead of taking advance courses in general stream a student can shift his/her centre of interest to any of the branches of Biology. These include Anthropology, Bioinformatics, Biomedical Engineering, Biotechnology, Computational Biology, Computer Simulation, Dairy Science, Environmental Management, Genetic Engineering, Medicine, Medical Transcription, Pathology, Surgery, etc.

Those who do not want to continue in the general and professional streams may opt for vocational courses, such as Agronomy, Apiculture, Breeding Biology, Forensic Science, Pharmacy, Pharmacology, Physiotherapy, Poultry farming, Prawn farming, Sericulture, and many others. The following table (Table 1.2) highlights some of the career options in biology.

Table 1.2 Career Options in Biology

Name of the Subject	Related Description
Agronomy	Production of crops and management of crop farms
Anthropology	Study of physical and mental constitution of man, cultural development of man and social evolution of human society since the remote past
Apiculture	Rearing and maintenance of bees, extraction and marketing of honey
Bioinformatics	Systematic development and application of computing systems and computational solution of techniques, analysing data obtained by experiments, modelling, database searching and instrumentation to make novel observations about biological processes
Biomedical Engineering	Production of artificial organs for human implants, artificial limbs, machines for artificial respiration (lung), circulation (heart), etc.
Biotechnology	Wilful manipulation, on a molecular level, of life forms or utilisation of knowledge pertaining to living systems.

Breeding Biology	Production of improved varieties of plants or animals; involves crossing of selected parents of the same or related species. It does not manipulate genes
Computational Biology	Systematic development, application and validation of computational hardware and software solutions for building simulation models of biological systems
Computer Simulation	Conversion of physiological phenomena into graphical and multi-dimensional and multimedia presentation without actually involving animals/plants
Dairy Technology	Rearing of improved varieties of cattle, such as cow, buffalo, etc., extraction, preservation and marketing of milk and milk-products
Environmental Management	Assessment of environment, finding out the ways and means for remedy of environmental problems and for conservation of biodiversity so as to maintain the balance of nature
Fishery	Culturing fishes in fresh water ponds and estuarine water, catching fish from fresh water streams, lakes and rivers and sea-water, including preservation, transportation and marketing
Forensic Science	Extension and application of scientific knowledge, including finger prints, blood typing, etc., to deal with criminal activities and laws
Genetic Engineering	Obtaining selected enes from an organism or synthesising copies of g and inserting them into another completely different organism
Medical Transcription	Interpretation and typewriting (transcribing) dictation from physicians and other healthcare provides regarding patient assessment and work-up surgical, radiology, and therapeutic of clinical course, diagonosis, and prognosis, etc.
Medicine	The science of treating diseases with drugs or curative substances
Microbiology	Study of structure, function, uses, effects and biological importance of microscopic organisms, including protozoans, algae, fungi, bacteria, etc.
Pathology	To deal with the nature of diseases, their causation, symptoms and effects
Pharmacy	The art or business of compounding, preserving and identifying drugs
Pharmacology	Related to the knowledge of action of medicines, their nature, preparation, administration and effects
Physiotherapy	Treating disability, injury and diseases by external physical means, such as electricity, heat, light, massage, exercise, etc.
Poultry Farming	Rearing of breeds of poultry birds like domestic fowl, chicken, ducks, their transportation and selling
Prawn Farming	Rearing, transportation and marketing of prawns
Sericulture	Rearing of different strains of silkworms, extraction of silk from them and transporting the reeled or spun silk for commercial purpose
Surgery	Related to anatomy and physical operations to cure the patients suffering from diseases that cannot be done with routine medicine

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SUMMARY

Biology deals with the science of living things — what they are, how they work, interact and change or evolve. For studying biology a scientific bend of mind is essential. Generally, the scientific method of gathering information involves some steps. These are Observation, Hypothesis Formulation, Hypothesis Testing and Developing Theories. Ancient Indians had some basic knowledge of biology as evident from such treatises as Susruta Samhita and Caraka Samhita. Although Aristotle classified animal species and arranged them into hierarchies, biology as a scientific discipline did not develop before the early sixteenth century. The most important early biologists were Vesalius and Harvey, who studied functional anatomy and Robert Hooke , who observed a thin slice of cork and recognised some honeycomb of empty, tiny 'microscopical pores' or compartments. Actual living cells were observed a few years later by Leeuwenhoek. Linnaeus introduced the system of classification and naming of plants and animals. Mendel discovered the laws of inheritance. Fleming discovered penicillin in his culture of the bacterium *Staphylococcus*. The penicillin found by Fleming was the toxic product of the blue mould *Penicillium notatum* that contaminated his culture of bacteria and was destroying the bacteria.

Cuvier was the first to identify the fossils of extinct bird-like reptile and laid the foundation of palaeontology. Lamarck first discarded the idea of 'fixity' of species. Schleiden developed the first statement on cell theory. His observation got support from Schwann. Darwin proposed natural selection as the mechanism of evolution of species. The concept of spontaneous origin of life was countered by Pasteur, who proposed the germ theory.

The works of Darwin, Pasteur, Mendel and many others have strongly founded the stage for the emergence of contemporary biology. With its development, biology has emerged as the most demanding of all sciences. James D. Watson and Francis H. C. Crick discovered the structure of Deoxyribose Nucleic Acid (DNA) in 1953 and this triggered a new era of molecular biology. W. T. Astbury, a British scientist, first used and defined the term 'molecular biology' in 1950. Har Gobind Khorana shared the Nobel Prize in 1968 with Robert W. Holley and Marshall W. Nirenberg "for their interpretation of the genetic code and its function in protein synthesis." Modern plant and animal breeders can now produce new forms of life with almost any mix of characteristics by altering the genes in DNA. Norman Ernest Borlaug won Nobel Prize in 1970 for his "green revolution", which involves the use of improved wheat seed, new types of higher-yield rice and more efficient use of fertiliser and water. Ian Wilmut and Campbell produced the first live, healthy sheep clone, Dolly, from fully differentiated adult mammary cells in 1996. In an attempt to reap the fruits of genetic engineering, the U.S. Department of Energy and the National Institute of Health has launched the U.S. Human Genome Project in 1990. The project aims to identify all the approximate 30,000 genes in human DNA and determine the sequences of the 3 billion chemical base pairs that make up human DNA.

Biology embraces a wide range of disciplines, such as zoology, botany and microbiology. The scientific classification of organisms, including their naming and identification is called Taxonomy or Systematics. Morphology is the aspect of biology that deals with the study of form, shape, size and structure of plants and animals. The study of morphology of internal systems or organs constitutes what is called Anatomy. Genetics unravel the mechanism of inheritance, maintenance of inheritance and cause of variation among organisms. The historical change in the properties of a population of organisms over generations is called organic evolution and this branch of biology is considered as Evolutionary Biology. Fossils in the earth crust represent the past history

of living beings and the study of fossils is called Palaeontology. Another approach in biology is to study the interrelationship between organisms and their environment, a branch known as Ecology. The study of the structure and composition of cells and tissues under the microscope is called Cytology and Histology, respectively. The branch of biology that deals with the mechanism of functioning of the life processes is referred to as Physiology.

Biology is in the forefront of human achievements, because of its manifold scope. Biology influences our daily lives and future. The development and use of modern medicine depends on the understanding of structure functions of cells, tissues, organs and organ-systems of the human body. Information about the life-histories of the causative agents of diseases and their relation with man are of much value for prevention and care of many diseases. Biology can help maintaining and increasing our treasures of medicinal plants. Also we can do away with many hereditary diseases and genetic disorders based on the understanding of genetics. It also helps in dispelling myths and disbeliefs.

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With modern biotechnology scientists have been able to produce genetically modified (GM) crops and successfully cloned animals and plants. The geneticists, evolutionists and ecologists can best judge the scientific and ethical aspect of such innovations based on biological background. Biology helps to understand how organisms interact with nature and the significance of maintaining biodiversity. It helps humans to assess their position in the world of life as a part of the earth's ecosystems. To make a good career in Biology, a student must pass 10+2 course with Biology, Physics, Chemistry and Mathematics. A student of biology can choose any of the related courses also.

EXERCISES

1. Define the term 'Biology'. Who proposed this term and in what sense ?
2. Make a distinction between biological and physical sciences ?
3. Give a schematic representation of the general steps involved in the methodical inquiries in science. Highlight the role of observation in the decision-making of a scientist.
4. What is a hypothesis ? Narrate the relation between observation and hypothesis.
5. What is an experiment? Highlight its utility in a scientific finding.
6. What do you understand by 'control experiment'? Explain how control experiment helped Louis Pasteur to put the concept of spontaneous origin of life to rest.
7. State the condition that leads to transformation of a hypothesis into a theory. Can every experiment lead to a theory? Give reasons in favour of your opinion.
8. Characterise the basic and applied researches and state their interrelation. Why a scientific finding needs to be published ?
9. Elucidate the connection between science and technology.
10. Fill in the blanks
    (a) *Chandyogya Upanishad* classified animals into _____, Andaja and _____.
    (b) *Susruta Samhita* classified all substances into _____and _____.
    (c) *Susruta Samhita* subdivided plants into Vanaspati, _____, _____, and _____.
    (d) According to *Susruta Samhita* and _____*Samhita* the two main divisions of animals are _____and _____.

(e) Indian medical tradition goes back to Vedic times when the _____ practised medicine and _____ practiced surgery on man.

(f) Susruta is acknowledged as the father of _____.

(g) The word _____ means the science of living or longevity.

(h) The constituents of Ayurvedic medicines are largely based on _____ matter.

11. Narrate the contributions of Aristotle in support of his talent.

12. Match the names given in Column I with items in Column II:

Column I	Column II
(a) Andreas Vesalius	1. Micrographia
(b) Antony van Leeuwenhoek	2. De Humani Corporis Fabrica
(c) Willium Harvey	3. Species Plantarum
(d) Carolus Linneaus	4. Philosophie Zoologique
(e) Robert Hooke	5. Anatomical exercise on the motion of the heart and blood
(f) Jean Baptiste Lamarck	

13. Give an outline of the post 1953 development of contemporary biology.

14. Briefly narrate the general trends of interdisciplinary approach in biological science.

15. Distinguish between

(i) Cell Biology and Molecular Biology.

(ii) Light microscope and Electron microscope.

(iii) Physiology and Biochemistry.

(iv) Biotechnology and Bioinformatics.

16. Write short note on Computer simulation and state its significance.

17. Elucidate the role of biologists in respect of

(i) Economic uses of plants and animals;

(ii) Usage of genetically modified food;

(iii) Understanding of the ecosystems; and

(iv) Human health and life.

18. Briefly describe the role of biology in dispelling myths, misconceptions and disbeliefs associated with the diseases like malaria or AIDS.

19. What is amniocentesis? Comment on the misuse of the technique of amniocentesis.

20. Write about any three career-options in biology according to your order of preference.

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Chapter 2

UNDERSTANDING LIFE

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You know that plants, animals, bacteria and fungi are living organisms whereas bricks, stones and rocks are non-living; viruses are neither living nor non-living. Also you might instantly recognise a living object and distinguish it from a non-living one. What makes something alive?

The dictionary meaning of life is the property that distinguishes living beings from non-living objects. Biologists, however, find it hard to define life, though they have vast knowledge of living things. It is difficult to draw a fine line between living and non-living things. We may support the above statement by taking an example of virus. The virus is a lifeless particle by itself, but it becomes active and multiplies rapidly when inside a living cell. Actually, life is not a definable entity or property. Rather than trying to define life precisely, biologists focus on how life works.

2.1 ORGANISMS SHARE SOME UNIFIED AND BASIC CHARACTERISTICS

Despite the extraordinary diversity, organisms have many things in common. Even the most diverse forms of organisms – fungi, plants, insects, and vertebrates – are built of cells that are similar in their internal structure and function. Starting from a single-celled microscopic amoeba or bacterium to the unique humans, all organisms share some unified and basic characteristics. These include order, energy utilisation, regulation or homeostasis, growth, development, reproduction, and adaptation. The basic features of living organisms are listed below:

(i) Highly organised and complex entities formed of one or more cells.
(ii) Carry out and control numerous chemical processes.
(iii) Acquire and use energy for metabolism.
(iv) Respond to changes in environment and maintain a constant internal environment.
(v) Grow in size, develop and produce offspring similar to them.
(vi) Adapt to environmental changes and gradually evolve into new types of organisms.

2.2 ANALYSING THE LIVING THINGS

Study of all living organisms shows that life has passed through simple to complex structural hierarchical levels, with each lower level emerging into a complex higher one. We now know that all organisms are formed of cells, either one or many. When a group of cells is meant for a specific function it constitutes a tissue. The tissues organise themselves into organs, which in combination with several organs, emerge as a system. We see that several systems are present and are required to perform diverse life processes in a multicellular organism. However, plants do not exhibit organisation exactly parallel to that of animals.

Levels of Biological Organisation

Biological organisation starts with submicroscopic **molecular level**, passes through microscopic **cellular level** and microscopic or macroscopic **organismic level** and ends in **ecosystems** and the **biosphere**.

The hierarchy of biological organisation (Fig. 2.1) reveals that atoms are the lowest unit at the molecular level while the cells are the smallest unit at microscopic level. Atoms combine to form molecules, which undergo chemical reactions to form organelles. Several organelles are contained in the cell. A group of cells meant for a specific function constitutes a tissue. Above the tissue level of organisation, many tissues

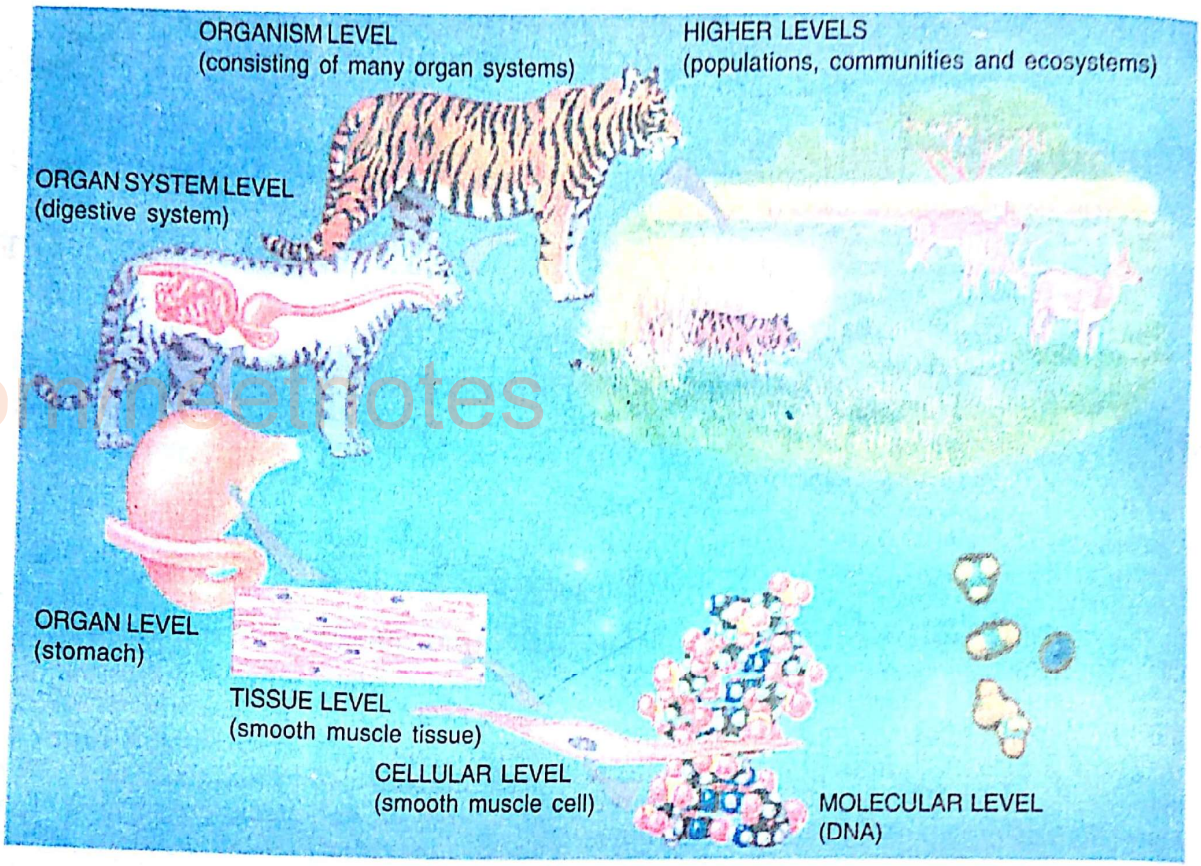

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Fig. 2.1 *The generalised hierarchy of biological organisation*

become engaged to form a particular organ, like liver, stomach, lung, testis, ovary, etc., for a specific function. A system emerges when many organs take up a particular life process, such as digestion, photosynthesis, respiration, reproduction and several others.

The living things are called organisms which, in turn, contain many individuals. Groups of individuals that interbreed or are potentially interbreeding form a species. These individuals form population. A localised group of organisms belonging to the same species constitutes a population. Populations of different species living in the same area make up a biological community. Community interactions, integrated with the non-living (abiotic) features of the environment, form an ecosystem. Within the ecosystem, an individual forms the smallest unit. A larger unit than ecosystem is the landscape, which is a geographical unit with a history. It shapes the features of the land and organisms that inhabit it. The entire zone of air, land and water at the surface of the earth that is occupied by organisms constitutes the biosphere.

What you would have understood from the above discussion is that we can identify the branches of science related to the different levels of organisation (Fig. 2.2). Thus, we need to look into the natural systems with a holistic approach and to see interconnectedness amongst the biotic and abiotic factors of environment.

Molecular Organisation : A Shift in Approach

Traditionally, our knowledge of living things is based on the observations made at morphological, anatomical and histological levels. No longer a biologist remains contented simply with morphology – structure of cells, tissues, organs, systems. Attention of biologists has shifted from organism level, i.e., the many ways in which living things differ from one another, to the molecular level, i.e., the way in which they are similar. In fact, the analytical techniques of physicists and chemists have enabled us to analyse the living things

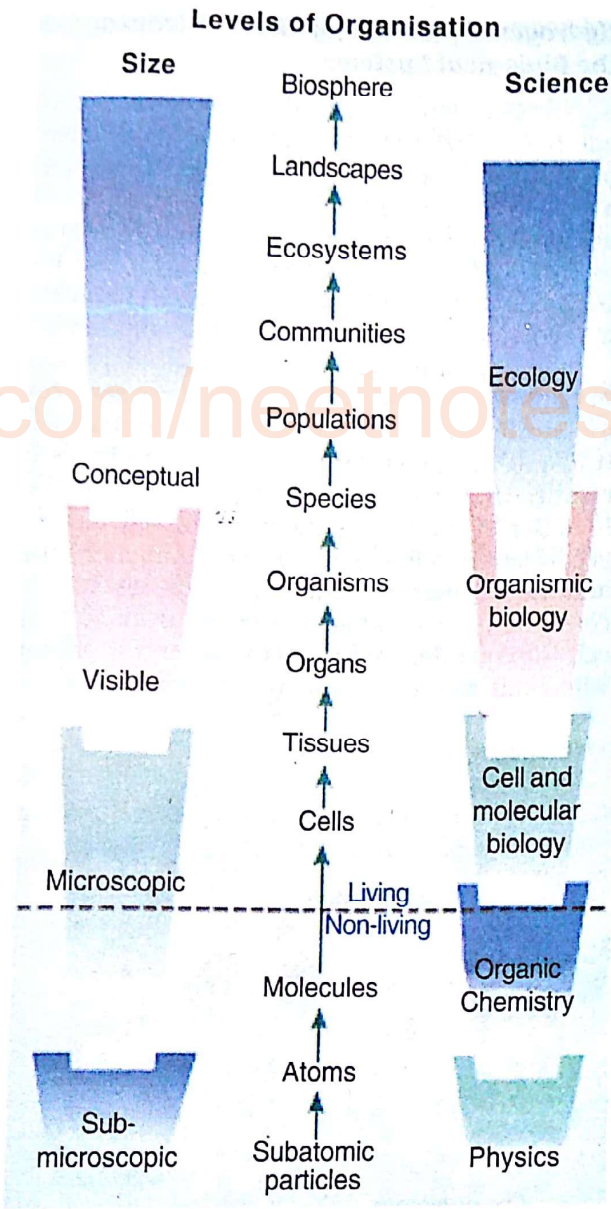

Fig. 2.2 *Various levels of organisation and the related branches of science*

at the level far smaller than microscopic level. Studies at the submicroscopic level have yielded a few prominent points. These are:

(i) The basic chemical organisations of living things are complex but remarkably similar.

(ii) The same materials and principles form the basis of both living and non-living things, that is, life is chemically based on the universal physico-chemical laws and obeys them.

(iii) Living matter lies in the biosphere interacts with its surrounding non-living lithosphere, hydrosphere and atmosphere.

Atoms are Nature's Building Material

All living plants and animals and the non-living substances, such as stones and rocks, are formed of matter. Any material in the universe that has mass and occupies a space is defined as matter. The building blocks of matter are **atoms**. Atoms aggregate and constitute **elements**. An element is a substance that contains only one type of atom. There are more than 100 elements occurring in nature. Of these, only 25 are essential to life. About 98 per cent of the mass of every living organism, be that a bacterium or a human being, is composed of just six elements, such as carbon (C), hydrogen (H), nitrogen (N), oxygen (O), phosphorous (P) and sulphur (S). Other elements present in small amounts in the living body are calcium (Ca), potassium (K), sodium (Na), magnesium (Mg), iodine (I), etc. Such elements that are required by an organism in minute quantities are considered as **trace elements**. They are of immense value as nutrients for the organisms. For example, the element iodine is necessary for the vertebrates as an ingredient of a hormone produced by the thyroid gland. Plants need molybdenum for incorporation of nitrogen into biologically useful substances.

Chemical Bonds are the Glue of Life

Atoms combine to form **molecules**. A molecule may be defined as two or more atoms linked by chemical bonds. A chemical bond is an attractive force that links two atoms to form a molecule. When two atoms share a pair of valence electrons, the bond is called **covalent bond** (Fig. 2.3). A bond in which a single pair of electrons are shared

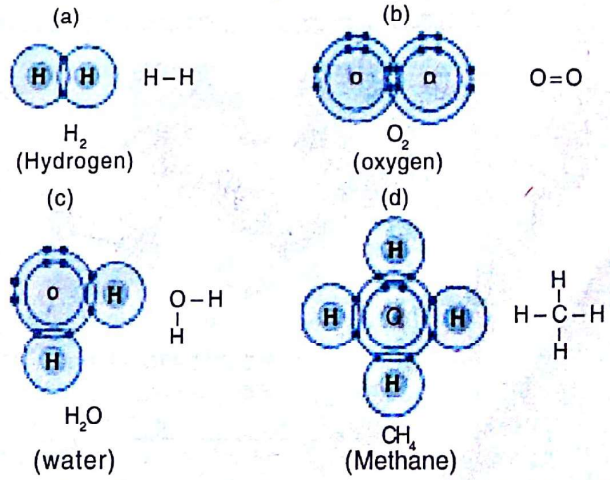

Fig. 2.3 *Covalent bonds (a) single bond (b) and (c) double bond (d) multiple bond*

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(e.g. H — H) is called a **single bond**. When two pairs of electrons are shared (O = O), the link is called **double bond.**

Water is a Polar Molecule

Molecules that exhibit charge separation are called **polar molecules** because of their magnetic poles. Water is a polar molecule. In the water molecule, the oxygen atoms bear a partial negative charge (δ^-) and each hydrogen atom a partial positive charge (δ^+) (Fig. 2.4a). In liquid water, the negatively charged oxygen atom of one molecule of water is attracted to the positively charged hydrogen atom of another molecule of water. The bond resulting from this attraction is called a **hydrogen bond** (Fig. 2.4b). Hydrogen bond is also formed when a positively charged (δ^+) hydrogen atom associates with one electronegative (δ^-) oxygen atom and at the same time becomes attracted to another electronegative (δ^-) sodium atom.

Hydrogen Bonds are of Great Advantage in the Biological Systems

A hydrogen bond is a weak bond; it has about one-tenth (10%) of the strength of a covalent bond between a hydrogen atom and an oxygen atom. Weak hydrogen bonds are of great advantage in the biological systems. The reason is that the contact between the molecules becomes very brief. The molecules separate immediately after reacting with one another.

Such advantage can be detected in the transmission of chemical signals to the receptor molecules of brain. The signal molecules transmit the signal by using weak bonds. As a result, they can separate much before the initiation of response in the receiving cell. If there is covalent bonding between the signal molecule and the receptor molecule, the receiving cell will continue to respond long after the transmitting cell stops sending message. Consider a situation when you continue to hear a call long after the

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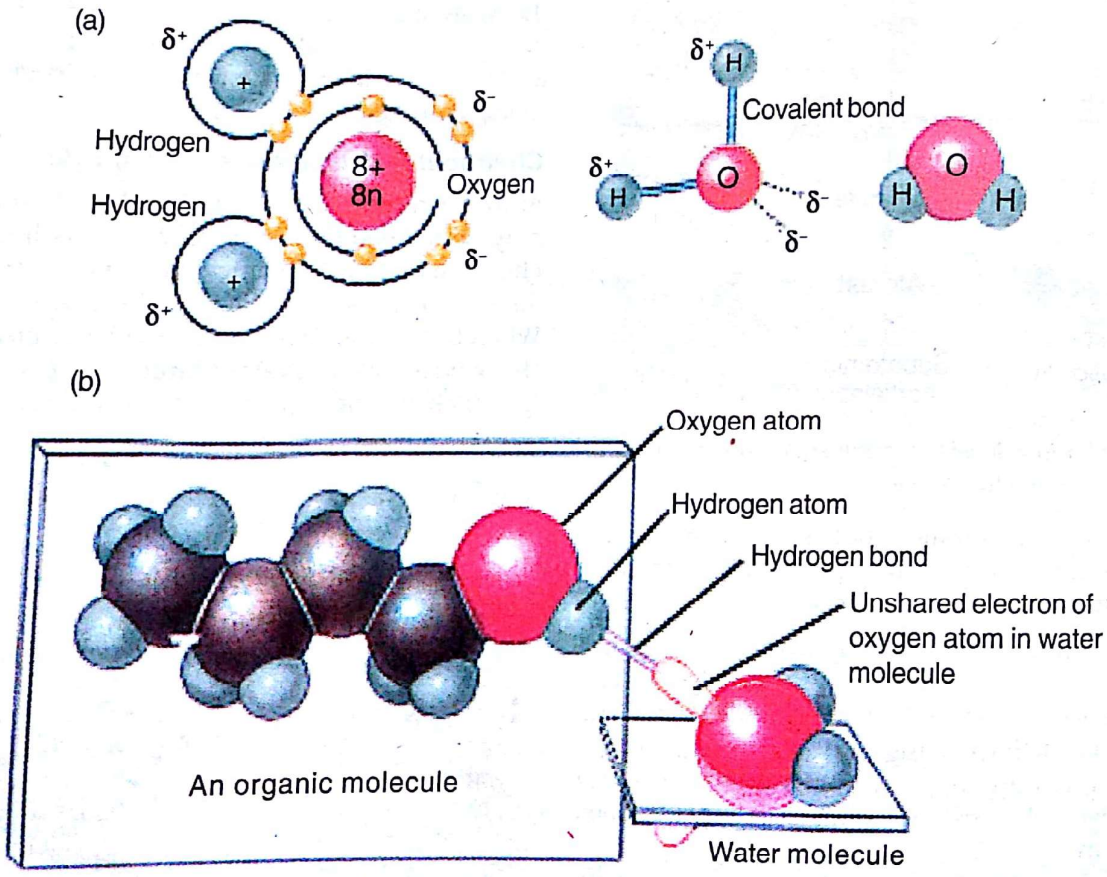

Fig. 2.4 *(a) Molecular structure of water (b) Hydrogen bond in water molecule*

caller has stopped calling. Nevertheless, when many hydrogen bonds are formed, they have considerable strength and greatly influence the structure and properties of substances. In fact, hydrogen bonding in water contributes to many of the properties that make water significant for living systems. They also play significant role in determining and maintaining the three-dimensional structure of giant molecules like proteins and DNA.

Ions Form Bonds by Electrical Attraction

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When one of the interacting atoms is much more electronegative than the other they cannot share their valence electrons. In such cases, two atoms are held together by a complete transfer of one or more valence electrons from the more electronegative atom to the more electropositive one. The atoms become charged and transform into **ions**. Ions are electrically charged particles or atoms that form when atoms lose or gain one or more electrons. The positively charged atom is called **cation** and the negatively charged one **anion**. The bonds formed by electrical attractions between ions bearing opposite charges (*viz.* cation and anion) are designated as **ionic bonds**.

tendency to join with another electron. Sodium atom can attain a stable condition, if the lone free valence electron is lost to another atom whose valence shell is deficient in electron. In the event of loss of this electron, sodium atom will be positively charged sodium ion (Na^+). Chlorine atom has 17 electrons – 2 in the inner energy level, 8 in the next level and 7 in the valence level. Again, this is an unstable condition. The addition of another electron to its valence level will make chlorine atom stable by transforming into a negatively charged chloride ion (Cl^-). Since the electronegativity of chlorine atom (3.1) is much higher than that of sodium atom (0.9), any electrons involved in bonding will tend to be much nearer to the chlorine nucleus. Consequently, when placed together, metallic sodium and gaseous chlorine react swiftly and explosively; sodium atom donates the free electron to chlorine atom, forming sodium and chloride ions (Na^+ and Cl^-). These oppositely charged ions remain associated in an ionic compound, sodium chloride, which is electrically neutral, and results into an extended lattice (Fig. 2.5). Hence, no direct sodium chloride (NaCl)

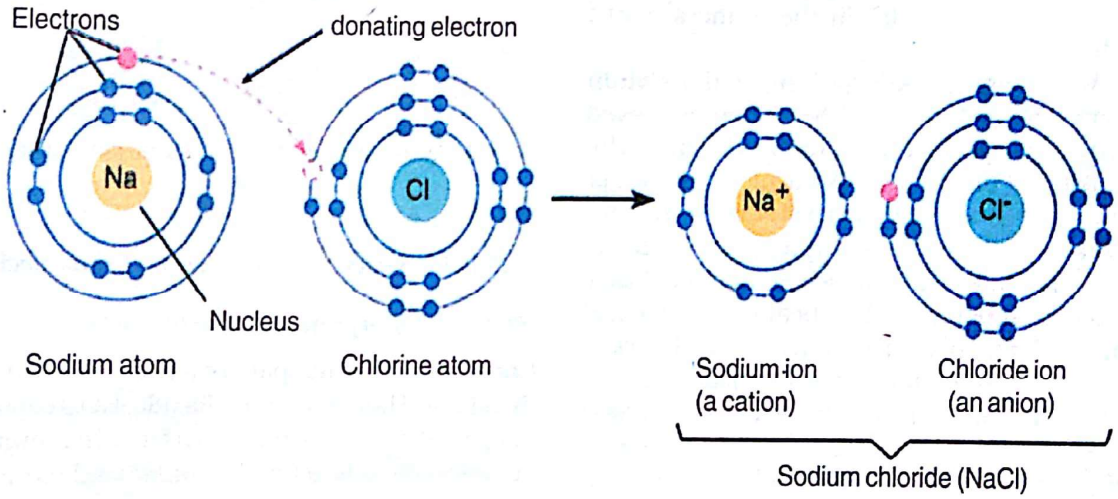

Fig. 2.5 *Electron transfer and ionic bonding in sodium chloride*

Common table salt, sodium chloride (NaCl), is an important lattice of ions in which the atoms are held together by electrical attractions (ionic bonds). Sodium atom has 11 electrons — 2 in the inner energy level, 8 in the next level and 1 in the valence level. The unpaired electron being unstable, has a strong

molecule is formed. The ions aggregate in a matrix with a precise geometry to form crystal. Such aggregations are called salt crystals.

Sodium chloride is an important constituent of our blood. Na^+ and Cl^- ions participate in the transport of materials through cell membranes.

Water Sustains Life

Water takes many forms (Fig. 2.6). Life on earth is totally dependent on water. Life originated in water and evolved there for about 3 billion years. Organisms require water-rich environment for their growth and reproduction. About two-thirds of our body is formed of water. Between 70 per cent and 90 per cent of living cells is water. All the building blocks of life, the organic molecules, including the monomers (amino acids, monosaccharides) and polymers (polynucleotides, polysaccharides) accumulated and interacted in water in the primeval earth conditions.

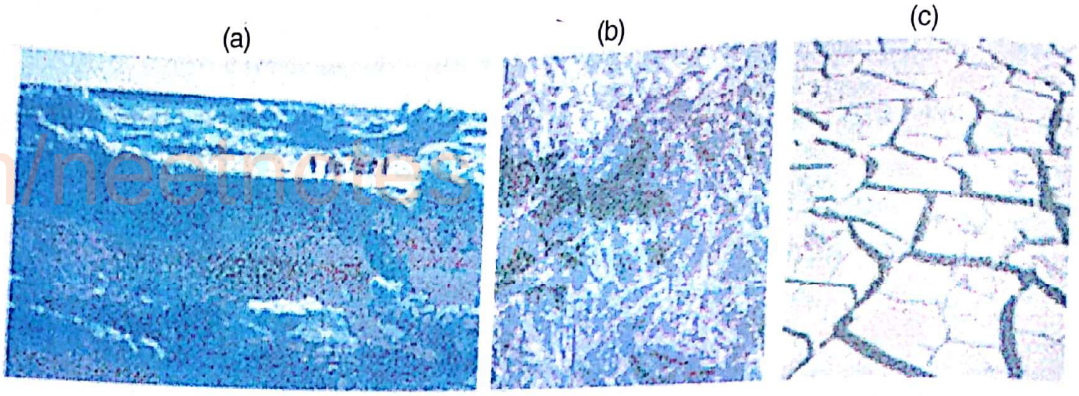

Fig. 2.6 (a) & (b) Water crystals of snow and ice (c) Scantiness of water

Why common salt and sugar dissolve in water? The polarity of water, as discussed earlier, underlies its chemistry and the chemistry of life. Being a polar molecule, water interacts with one another. Water is an effective solvent because it is capable of forming hydrogen bonds. Water molecules gather closely around a substance that bears an electrical charge. When placed in water, the electrical attraction of water molecules disrupts the forces holding the Na^+ and Cl^- ions in their crystal matrix of NaCl. This results in the dissociation of the NaCl crystals into Na^+ and Cl^- ions. Then the water molecules form hydrogen bonds around individual Na^+ and Cl^- ions and form a **hydration shell** around each ion (Fig. 2.7). Consequently, the ions cannot reassociate by forming ionic bonds.

The same explanation can be advanced as to why sucrose (table sugar) dissolves in water. Sucrose is composed of molecules that contain slightly polar hydroxyl (OH) group. Water molecules form hydrogen bonds with individual OH groups of the sugar molecules. Actually, the large number of hydrogen bonds, so formed, are responsible for many of its properties. These include cohesion, high specific heat, and high rate of vaporisation, good solvent for other polar molecules, storing of heat and maintenance of relatively constant pH in the living systems.

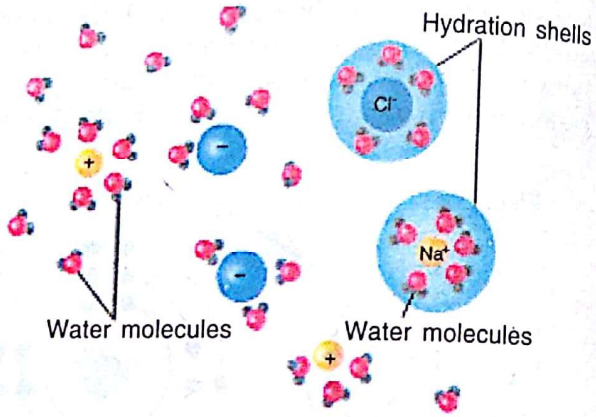

Fig. 2.7 Water molecules form hydration shells

Principal Inorganic Source of Carbon

Carbon is the principal structural element of a living cell. Though carbon dioxide (CO_2) contains carbon (C), it is simpler than other organic compounds. It is generally considered inorganic and is the principal inorganic source of carbon. Our atmosphere contains only about 0.033 per cent carbon dioxide (CO_2). Before it can take part in a chemical reaction, CO_2 needs to get dissolved in water. The thin aqueous films covering almost every cell is sufficient to dissolve CO_2. The dissolved CO_2 then reacts with water and forms carbonic acid. CO_2 and H_2O are the raw materials from which plants produce many complex essential organic compounds.

Molecular Oxygen is Necessary for Life

Molecular oxygen (O_2) constitutes approximately 21 per cent of the atmosphere. It is essential for life and is utilised by most plants and animals in the process of extracting energy from the nutrients. Oxygen serves as the ultimate acceptor of electrons. In its absence, the cells exhibit only 5 per cent of their normal efficiency. Oxygen is weakly soluble in water. But the small amount of dissolved oxygen is ample to meet the needs of the aquatic organisms, provided the water surface is exposed to air. Alternatively, the water body must hold aquatic plants. In fact, the green plants release oxygen during photosynthesis. This oxygen is the source of all atmospheric molecular oxygen.

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Building Blocks of Life

Oxygen, carbon dioxide and water are truly basic to life, but the organisms also require many other compounds to capture, store, transport and utilise energy to sustain life. Organic chemistry includes the reactions that are critical to life.

Organic molecules may be small or large. They contain one or more functional groups — simple sugars or monosaccharides contain hydroxyl (–OH) and CHO or C = O group. Large and complex assemblies of simple biological molecules are called macromolecules. For example, linking of many simple ring-shaped sugars or monosaccharides results in the formation of polysaccharides. Similarly, several amino acids form proteins; several nucleotides produce polynucleotides (DNA and RNA). From this, we can consider the macromolecules as polymers of small and simple molecules. Macromolecules are traditionally grouped into four major categories: carbohydrates, lipids or fats, proteins and nucleic acids (Fig. 2.8).

Carbohydrates — the Main Energy Storage Molecules

The simplest units of carbohydrates are the monosaccharides, for example, Glucose $(CH_2O)_6$ or $C_6H_{12}O_6$. Two such monosaccharides may join by covalent bond and form 'double sugars' or disaccharides (e.g. sucrose, a sugar; lactose, a milk sugar). Simple sugars or monosaccharide subunits join together to form polysaccharides, such as starch (the principal carbohydrate storage product of higher plants), cellulose (the major supporting material of plants), chitin (the major structural component of insect exoskeleton and fungal cell walls) and glycogen (the principal carbohydrate storage product of animals). Carbohydrates contain either –CHO or C = O bonds in which they store energy and function as energy storage molecules. This energy is released when the bonds are broken. Besides, carbohydrates make up the body of organisms and serve as carbon skeletons that can be rearranged to form other molecules important for biological structures and functions.

Lipids — A Major Group of Insoluble Hydrocarbons having Many Functions

A simple form of lipids are hydrocarbons with a carboxyl (–COOH) group at one end. Being nonpolar, such lipids are hydrophobic and insoluble or partly soluble in water. Lipids dissolve in organic solvents like ether. Most lipids

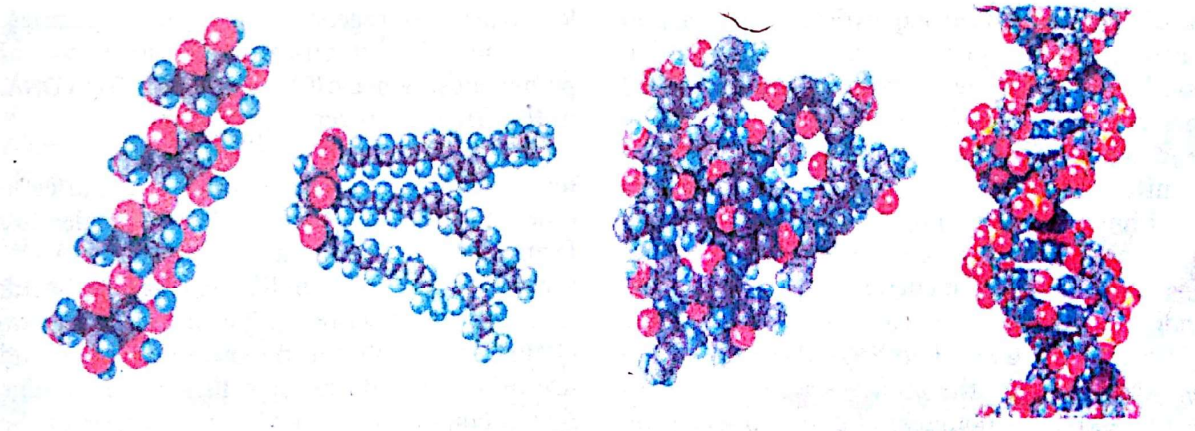

Fig. 2.8 Biological macromolecules

have an ionic group attached to the carboxyl end. The best example of lipids is the neutral fats, called oils when liquid. A fat molecule is formed of fatty acids held together by a backbone of three carbon atoms, each carrying a hydroxyl (–OH) group. Fats store energy and provide insulation, cushioning and protection to the body. Lipids may also contain elements like phosphorous and nitrogen. The membranes of cells are formed of modified lipids called phospholipids which contain one unit of glycerol, two units of fatty acids and a phosphate group (PO_4) at one end, often linked with a nitrogen-containing group. The phosphate group is polar and hydrophilic, whereas the other end, being formed of two long chains (hydrocarbon tails) of fatty acids, is nonpolar and hydrophobic. In water, the nonpolar tails of phospholipids aggregate away from water, forming a lipid bilayer. This bilayer is the basic framework of biological membranes.

Membranes often contain **steroids**. This is a type of lipid that contains four carbon rings. The steroid cholesterol is present on most animal cell membranes. Other kinds of steroids are hormones like testosterone and estrogens. Terpenes, component of many biological pigments (e.g. chlorophyll in plants and the visual retinal pigment of animals), are long chain lipids. Rubber is also a terpene. Prostaglandins are formed of 20 carbons that are modified fatty acids. Prostaglandins act as local chemical messenger in many vertebrate tissues.

Proteins : Structural and Functional Make Up of Cells

All proteins are formed of subunits called amino acids. Only 20 different amino acids, which are made primarily of carbon, hydrogen, oxygen and nitrogen, are required for proteins. An amino acid is a molecule containing amino ($–NH_2$) group, a carboxyl (–COOH) group and a hydrogen (H) atom, all bonded together to a central carbon (C) atom. Besides, each amino acid has a side chain or group. The side chains may be simple (e.g. glycine) or complex (e.g. tryptophan). It is the chemical properties of the side groups of amino acids that determine the types and functions of proteins. If the side group is polar or ionic, the amino acid is soluble in water and if it is nonpolar at a pH 6.5 and 7 the amino acid is insoluble in water.

The amino acid building blocks link together by covalent bonds called peptide bonds. These bonds form a chain of amino acid units which is called polypeptide chain. Protein molecules often consist of more than one polypeptide chain. The chains may be held together by weak hydrogen bonds (e.g. the protein haemoglobin), both hydrogen and covalent bonds (the protein hormone insulin).

Proteins perform many functions. They make up the main structural and functional components of cells. About 50 per cent of the dry weight of living matter are protein. Most organisms have between 1000 and 50,000 proteins. Also they form structural elements like collagen (which forms matrix of bone cells, skin, etc.) and muscle proteins like actin and myosin (which play the key role in muscle contraction). Biological catalyst or enzymes (e.g. hydrolytic enzymes, which cleave polysaccharides), hormones (e.g. insulin, which controls blood sugar), immunoglobulins (e.g. antibodies, which mark foreign proteins for elimination) and transporters (e.g. haemoglobin, which transports oxygen and carbon dioxide in blood) are nothing but proteins.

Nucleic Acids : Information Storage Devices of Cells

The nucleic acids are linear polymers of repeating monomer subunits called **nucleotides**. Each nucleotide is formed of a pentose sugar, a phosphate group and a nitrogenous base (purine or pyrimidine). The sugar is ribose in ribonucleic acid (RNA) and deoxyribose (lacking one oxygen atom) in deoxyribonucleic acid (DNA). There are four types of nitrogenous bases, two of purines, adenine (A) and guanine (G) and two of pyrimidines, cytosine (C) and thymine (T) in DNA. In RNA thymine is replaced by uracil (U).

The sequence of nucleotides in a DNA molecule is constant for a species but varies in different species. Usually, DNA molecules are formed of two polynucleotide chains (double-stranded) that are held together by weak hydrogen bonds between adjacent bases. The two chains are oriented in opposite directions and are arranged side by side like a ladder. The nitrogenous bases form the cross-rungs of the ladder. Above all, the entire double-stranded

structure becomes coiled spirally or helically around central axis transforming into a double helix. Most RNA molecules consist of one polynucleotide chain (single- stranded).

2.3 THE ENERGY TRANSFER DEVICES OF LIFE

Cellular activities such as growth, movement and active transport of ions across the cell membrane require energy. No cell manufactures energy but all organisms take in energy and transform it into another kind to do many kinds of work. Green plants and bacteria take in solar energy to produce their own chemical energy (food). Animals obtain food from outside and break it to obtain energy for performing the different physiological functions. The potential for work is present where there is energy to be tapped. It can be weight of water stored behind a dam, covalent bonds of glucose, an electron excited into a higher orbital by sunlight or tightly bound nuclei of nuclear plants.

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Energy is often Transferred with Electrons

All atoms possess energy. It takes work to keep the electrons in the orbital of an atom. Virtually, all the energy for living organisms comes as radiation in the form of photons from the sun and is captured by electrons. Organisms reap and use this energy to fuel all the processes of life. You have already learnt that during chemical reactions electrons are transferred from one atom to another. The loss of an electron is called oxidation, whereas the gain of an electron is regarded as reduction. Reduction-oxidation **(redox)** reactions play key role in the flow of energy through biological systems.

What is Enthalpy and Free Energy ?

In a biological system, the total energy, including usable energy that can do work and unusable energy that is lost to disorder is called **enthalpy**. The amount of usable energy that is available for doing work when temperature and pressure are uniform throughout the system is the **free energy**. It is 'free' because it is available for work under certain conditions.

The energy required to destabilise chemical bonds and to initiate a chemical reaction is called **activation energy**. Reactions that occur without outside intervention, release free energy and can perform work, are called **spontaneous reactions**. Energy yielding chemical reactions are known as exergonic reactions. On being initiated by activation energy such reactions take place spontaneously. A chemical reaction to which a net input of free energy, from outside source is needed for its initiation is called endergonic reaction. An exergonic reaction has an endergonic first step because, for two molecules to combine, they must come unusually close and frequently one or more pre-existing bonds must break.

How Energy Flows

In each step of the hierarchy of biological organisation, chemical reactions play the intricate and key role by a constant flow of energy. All the daily activities, like running, walking, moving, etc. require change in the form of energy. The **Laws of Thermodynamics** describe how energy changes. Thermodynamics is the study of energy transformations that occurs in a collection of matter. According to the **First Law of Thermodynamics**, the total amount of energy in the universe is constant. Energy can change from one form to another but it can never be created or destroyed.

If the energy needed to do work in a cell or a particular system is not already available internally, it must be obtained from a source outside the system, which thereby loses a corresponding amount of energy. When an organism takes food it actually acquires energy by transferring some of the potential energy stored in the food to its own body. A green plant traps solar energy and converts it into high-energy adenosine triphosphate (ATP) molecules. This energy is converted to potential chemical energy in the form of carbohydrates (food molecules).

$$12H_2O + 6CO_2 \xrightarrow{\text{solar energy}} \underset{\text{(sugar - energy rich compound)}}{C_6H_{12}O_6} + 6O_2 + 6H_2O$$

However, within a living system the potential chemical energy may be converted into other forms, such as kinetic energy (a measure for random motion of molecules) in the form of light or electricity. During such conversions some of the energy diffuses in the environment as heat. The **Second Law of Thermodynamics** explains why the total amount of free energy is declining in the universe as a whole. This

law states that every transfer or transformation of energy makes the universe disordered. In other words, no physical process or chemical reaction is 100 per cent efficient. Some energy is lost to associated disorder. Scientists use a quantity called **entropy** as a measure of the disorder or randomness. Entropy of the universe increases with every transfer or transformation of energy. The potential energy that the universe held after its origin some 10 to 20 billion years ago would never be available. Why this increasing entropy of the universe is less apparent? This is so because the entropy takes the form of increasing heat, which is the energy of random molecular motion.

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Organisms are Open Systems

If energy can never be destroyed (First Law of Thermodynamics), what prevents the recycling of energy ? Answer to this question can be found in the Second law of Thermodynamics. The term 'system' denotes any part of the universe containing specified matter and energy and whose energy transformations are studied in thermodynamics. The rest of the universe remains outside the system and is considered as surroundings. In an open system, such as living cells, matter and energy can be transferred between the system and the surroundings. The very term 'open' refers to the fact that there is exchange of materials and energy between the organisms and their surroundings. Organisms are **open systems** (Fig. 2.9a) because they interact continuously with their environment.

A closed system is one that does not exchange matter and energy with its surroundings. For instance, a warm liquid is placed in a thermos flask or a bottle and the lids are tightened. Is it not the fact that the liquid has been isolated from its surroundings? The thermos flask more or less represents a **closed system**. (Fig. 2.9b). There is no question of exchange of matter between the internal environment of the thermos flask and its surroundings. So far as energy is concerned, the liquid remains warm for considerable period of time, depending upon the efficiency of the thermos flask. During this period, the heat energy of the liquid is not released outside and it moves inside. This keeps the liquid warm. Hence, thermos flask represents an example of closed system. There is no reason to believe that the first law of thermodynamics does not apply to living organisms. In fact, it applies to the universe as a whole or to any closed system in the universe. It should be remembered that an open system is only a part of a larger closed system.

2.4 METABOLISM IS THE TOTAL OF ALL CHEMICAL REACTIONS

Life on earth involves a never- ending flow of energy within cells– from one cell to another, and from one organism to another. All living organisms need a continuous supply of energy and materials. Exchange of matter and energy between an organism and its environment and the transformation of this matter and energy within the organism is called **metabolism**.

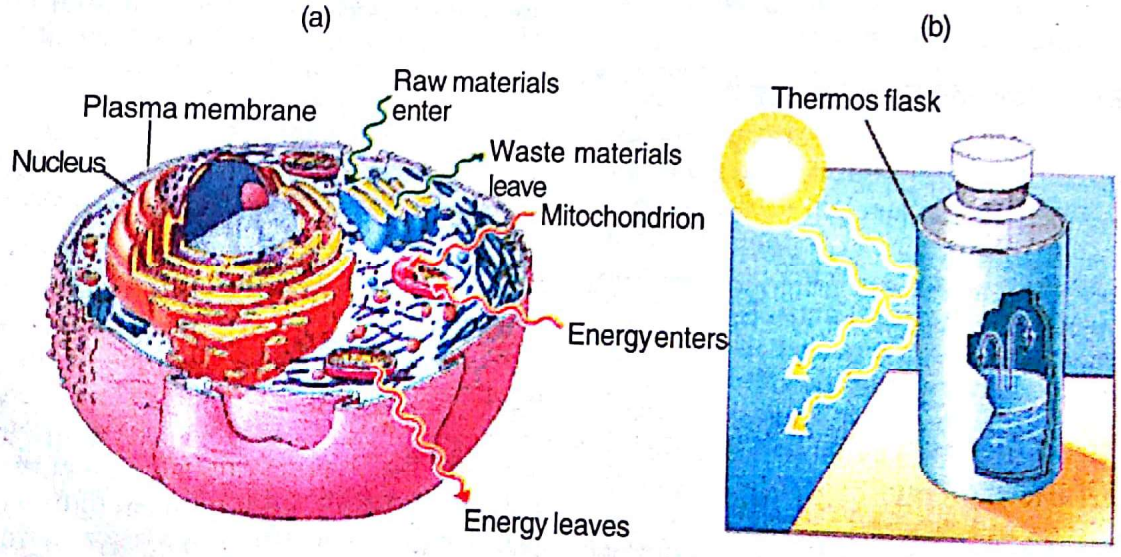

Fig 2.9 (a) Open System (b) Closed System

In other words, metabolism is the total of all chemical reactions carried out by an organism and arises from specific interactions between molecules within the interior of cells. The metabolic reactions of all cells are unusually similar despite the enormous differences in the cells and organisms.

Enzymes Direct Metabolic Pathways

A substance that speeds up a reaction but itself remains unchanged at the end of the reaction is a catalyst. An inorganic catalyst like platinum is relatively unselective about the reactions it helps. But the cells require specific catalysts for specific reactions. For this reason, cells have evolved an enormous variety of highly specialised organic catalysts, called **enzymes**.

Organisms contain thousands of different kinds of enzymes that direct a variety of reactions. Many of these reactions in a cell take place in sequences called biochemical pathways. Enzymes guide the biochemical pathways along desired directions. Most enzymes are globular protein molecules with one or more pockets or clefts on their surface called active sites. A particular enzyme generally interacts with only one type of reactant or a pair of reactants called the substrate. Substrates bind at the active sites of the enzymes forming enzyme-substrate complex. For example, the enzyme sucrase will act only on sucrose to produce glucose and fructose. It will not act on other disaccharide, such as maltose.

$$\text{Substrate} \xrightarrow{\text{Enzyme}} \text{Product}$$

$$\text{Sucrose} + H_2O \xrightarrow{\text{Sucrase}} \text{Glucose} + \text{Fructose}$$

As a whole, metabolism is concerned with managing the material and energy resources of the cell. Some metabolic pathways release energy by breaking down complex molecules to simpler forms. These reactions harvest energy by breaking chemical bonds and are regarded as catabolic pathways or catabolism. In cellular respiration, glucose ($C_6H_{12}O_6$) and other organic compounds are broken down to produce CO_2 and H_2O and to release energy.

$$C_6H_{12}O_6 + 6O_2 \xrightarrow{\text{Enzymes}} 6H_2O + 6CO_2 + \text{energy}$$

(Glucose)

Catabolic pathways
(Cellular respiration)

$$12H_2O + 6CO_2 + \text{Light energy} \xrightarrow{\text{(Chlorophyll+Enzymes)}} C_6H_{12}O_6 + 6O_2 + 6H_2O$$

(Glucose)

Anabolic pathways
(Photosynthesis)

In the reverse situation, reactions (e.g. photosynthesis) consume energy to make complex molecules, like glucose from simple forms, like CO_2 and H_2O. This is called anabolic pathways or anabolism.

Biochemical Pathways are highlyRegulated

What will be the result if both anabolism and catabolism occur simultaneously? Apparently, the net result can be guessed as to be a chemical chaos. The reason is very simple. A substance synthesised by anabolism would be immediately broken down by catabolism. On the other hand, synthesis of an excess amount of compound in a biochemical pathway would result in the unnecessary wastage of energy and raw materials. In effect, there is no such scope for either chemical chaos or wastage of energy and raw materials to occur. Actually, operation of each metabolic pathway is tightly under the control of cell's regulatory systems. Enzyme activity is sensitive to the presence of specific substances that bind to the enzymes. A substance that binds to an enzyme and decreases its activity is called an inhibitor; if it increases the activity of an enzyme it becomes an activator.

The regulation of simple biochemical pathways often depends on the allosteric site on the enzyme that catalyses the first reaction in the pathway. An allosteric site is the part of an enzyme, away from its active site, that can switch on/off the enzyme's function. In the metabolic pathways, the product of one reaction may serve as the substrate for the next reaction. Hence, enzymes ordinarily route matter through the biochemical pathways by selectively accelerating each step. When necessary, the final end product of one pathway may become the **allosteric inhibitor** for the action of the first enzyme of that pathway. This method of regulating mechanism is called **feedback mechanism** (Fig. 2.10). Thus the regulation of simple biochemical pathways often depends on the feedback mechanism.

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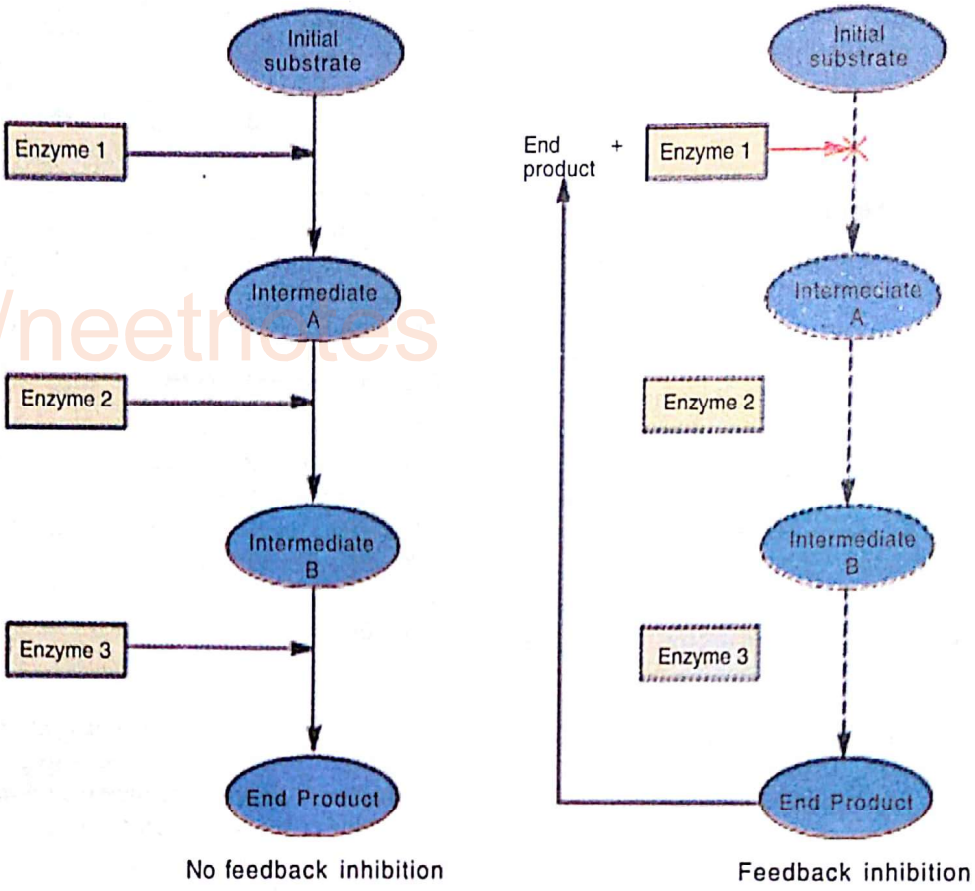

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Fig. 2.10 *Biochemical pathways*

2.5 HOMEOSTASIS – A FUNCTION OF REGULATORY SYSTEM

All organisms maintain relatively constant internal conditions different from their environment. For this, many organisms have regulatory mechanisms, which co-ordinate their internal functions, such as providing the cells with nutrients, transporting substances through the body. Others simply adjust to the environment by adopting the temperature, salinity and other physical aspects of their surroundings. In any case, maintenance of a dynamic constancy of the internal environment or steady state is termed as **homeostasis**.

Organisms are also capable of adjusting with the fluctuation of their external environment. Similarly, cells of the multicellular organisms have the ability to regulate their extracellular fluid that bathes them. They can maintain the concentrations of glucose, oxygen, carbon dioxide and water and ions of sodium, calcium, hydrogen, etc. in the intercellular matrix within narrow limits. The cells also respond to signals, such as growth factors, hormones, etc., in their immediate environment. In doing so they participate in regulating the body as a whole.

Homeostasis is a fundamental property of life. It is considered to be a sign of good health. That is why the physicians commonly tell us to keep record of body temperature, test blood composition and pressure and other measures of internal environment whenever we fall ill.

Thermoregulation

In the living cells, thermal energy (heat) is produced during exergonic reactions of metabolism. The vast majority of vertebrates (fishes, amphibians, reptiles) and plants lose most of their thermal energy to their environment. Such animals are called

ectothermic. These organisms often depend on their environment for temperature regulation. Their skin may appear as either cold or warm, depending on the environment from which they have been taken for examination. If any of them is picked up from a hibernaculum or caught beside a cold body of water, it is likely that the skin will be cold. This is the reason for calling them as **poikilothermous**. Alternatively, if an animal is examined during warm afternoon while it was basking in the sun, its skin will appear warm. Actually, these animals save energy for utilising in other functions like reproduction.

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Mammals, birds and a few fishes (e.g., tuna fish and sword fish), instead of losing thermal energy, actually retain it for use. They have developed insulating devices, such as fat, hair, feathers, etc., for retardation of heat loss to the environment. They are **endothermic**. Their body temperature remains relatively constant, which is usually higher than that of the environment. For this reason they are called **homeothermous**. Any change in external temperature will not affect their body temperature. When the ambient temperature is cold, superficial blood vessels constrict (vasoconstriction) to divert the warm blood to deeper vessels. When the external temperature is warm, the superficial vessels dilate (vasodilation) so that the warmth of the blood can be lost by radiation.

Homeothermy in Humans

Being a mammal, we are endothermic and homeothermic as well. To maintain the body temperature, which is 37° C (98.6° F), we have sensors that detect the set point. This can be compared with the functioning of the thermostat of a room air-conditioner machine. Usually, the temperature of the thermostat is set at 21°C (70° F). If the temperature of the room rises sufficiently above the set point, a sensor present within the thermostat detects the change and activates the effector of the machine. The air-conditioner then reverses the deviation from the set point of the thermostat. Finally, the room temperature is controlled at the set point of the thermostat.

Our skin contains two types of sensory neurons. They are sensitive to changes in the temperature outside our bodies. They are termed as **thermoreceptors**. Some of them are sensitive to cooler temperatures and are called **cold receptors**, while the others are sensitive to warmer temperatures and are called **warm receptors**. The former set is stimulated by fall in temperature and the latter by the rise in temperature. Conversely, warming inhibits cold receptors and cooling inhibits warm receptors. The warm receptors are located immediately below the epidermis (skin), while the cold receptors are located slightly deeper in the dermis. We have thermoreceptor also within the hypothalamus of our brain. These receptors monitor the temperature of the circulating blood and send the information of the body's internal (core) temperature to the brain.

The thermoreceptors of the skin sense, say for example, any rise in external temperature caused by a perturbing factor like the sun, and send the message to the thermoreceptors of the hypothalamus, which detect the change in the accompanying body temperature. The hypothalamus now responds by activating the effector, that is, the sweat glands and the blood vessels of the skin. As a result, release of sweat and dilation of the blood vessels are initiated, which cause cooling and diffusion of body temperature. In other words, the set point is defended by the hypothalamus. Since the regulation of temperature is by cooling the body, in this case, it is in the negative side or reverse direction. This type of control system is called **negative feedback loop** (Fig. 2.11).

When the perturbing factors of temperature are snow or ice, the hypothalamus induces the blood vessels of our skin to constrict and skeletal muscles to contract and we shiver. In that case, the temperature is not lost due to radiation but raised due to blood going to deeper vessels. Thus, negative feedback loop keeps the body temperature within a normal range.

2.6 GROWTH, DEVELOPMENT AND REPRODUCTION

All organisms grow, develop and reproduce. Increase in mass or overall size of a tissue or organism or its parts is called growth. Growth occurs due to synthesis of two different kinds of substances. These are protoplasmic substances, such as cytoplasm and nucleus, and apoplasmic substances, such as fibres of connective tissues, matrix of bone marrow and cartilage. **Apoplasmic substances** are those substances that are produced by the cells and they form a constituent part of the tissues. Secretions of cells like digestive juices, sweat, etc. are removed from the cells and organisms. As such these do not fall in the category

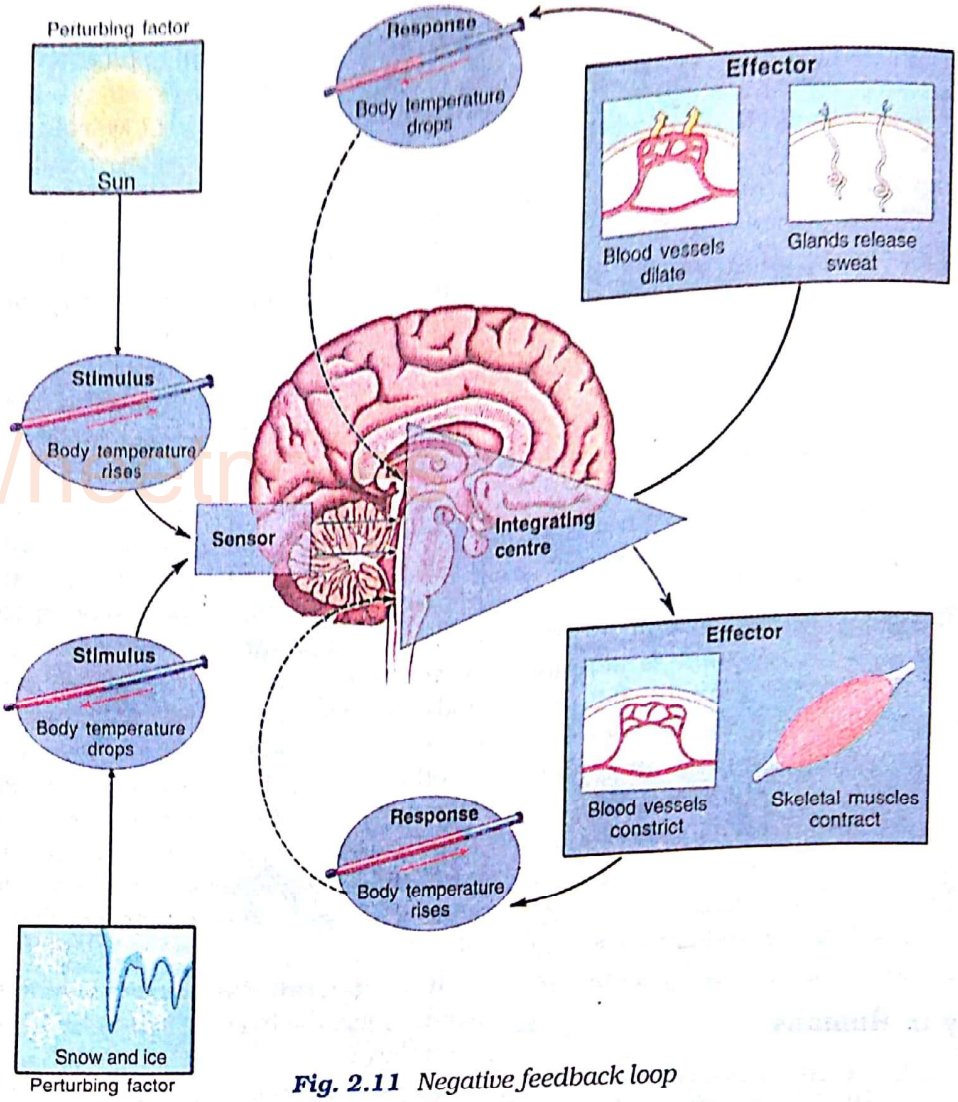

Fig. 2.11 *Negative feedback loop*

of apoplasmic substances. Similarly, we take water and food and our body mass increases. Such increase in body mass cannot be considered as growth.

Growth is the result of metabolism, which involves the transfer of energy. When the rate of anabolism equals to that of catabolism, no increase in bulk of the cells or organism occurs. If the rate of the synthetic process or anabolism exceeds that of the destructive process or catabolism, growth occurs. In the reverse situation, there will be degrowth.

In general, growth involves three processes or strategies. These are cell proliferation, cell enlargement and secretion of large amount of extracellular matrix. Growth is one of the three important characteristics of development. This is revealed by the fact that cell proliferation, cell enlargement and secretion of extracellular materials occur during development of a zygote into an embryo.

The other two features that characterise development are **morphogenesis** and **differentiation**. Morphogenesis produces new forms by involving cell movements. For example, a zygote transforms into blastula, a blastula into a gastrula and a gastrula into a miniature adult. Differentiation results in increasing diversity of cells. Some cells form nervous system, some others form heart and circulatory system and so on. Cells of the nervous system are different both in structure and function from those of circulatory system. Attainment of such structural and functional diversity is the main feature of differentiation.

Reproduction involves the production of progeny of the organisms. Reproduction is necessary for the continuation of life and to compensate for the loss of life due to death. Organisms reproduce by asexual or sexual means. In asexual reproduction the individuals

undergo binary or multiple fission to produce two or many offsprings. Sometimes a part of the body can develop into an adult by fragmentation or vegetative reproduction. Sexual reproduction involves the formation of specialised sex cells, called spermatozoa in males and ova in females. All organisms pass their hereditary materials to their offspring during reproduction.

2.7 ADAPTATION

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You might know that birds have wings, spiders design web, night flowers are usually white and emit scent, desert plants are either leafless or have fleshy leaves and succulent or juicy stems and so on. Usually, the beak of humming birds is extremely narrow and elongated and they suck nectar from flowers that are usually red or yellow, nearly odourless and lack a landing platform (Fig. 2.12).

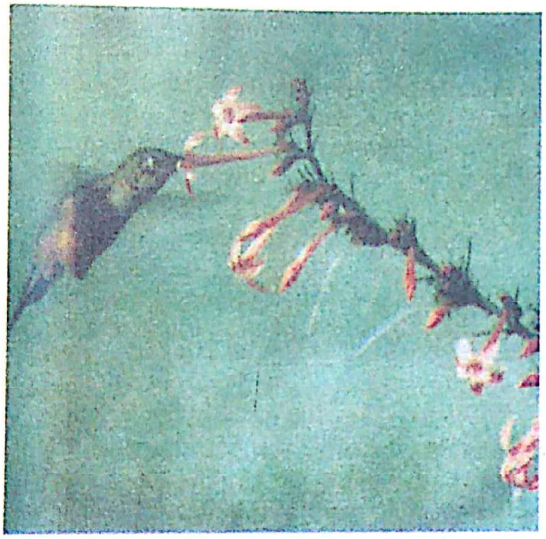

Fig. 2.12 A humming bird hovering around scarlet gilla

You might see that some species of orchids resemble in shape, the colour and odour of the females of certain species of bees or flies (Fig. 2.13).

Are these features of animals and plants without any significance? What is the common point that can bind all these different characteristics together? Do they have any biological significance? A close examination and analysis of the characteristics of organisms would offer an explanation for linking the different features. Birds use the wings for flight, spiders design web for trapping their prey such

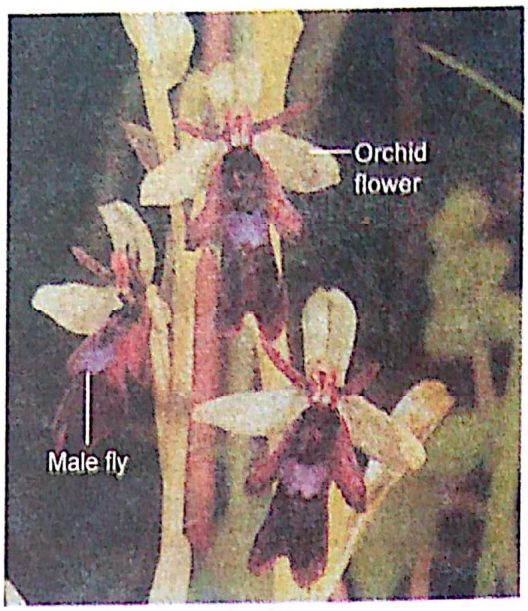

Fig. 2.13 An Orchid that assumes the shape of a female fly to attract male flies

as flying insects, night flowers are so to attract pollinators and desert plants are so for conservation of moisture. Taking the example of humming birds, we can explain that these birds suck nectar from some flowers without landing on them because there is no platform to land. Humming birds can suck nectar while hovering in front of flowers because of their extremely narrow and elongated beak. Likewise, orchids assume the shape of the females of certain bees or flies and the male bees of that species mistakenly attempt to copulate with orchid flowers. However, in doing so, they transfer pollens from flower to flower.

What you can conclude from the analysis of the above examples is that adaptation to the environment is one of the most distinctive features of living beings. **Adaptation** may be defined as any characteristic of an organism, which makes the organism better suited to its environment. An organism is best adapted to a given environment when it possesses inherited features that enhance its survival and ability to reproduce in that environment. Adaptation may be structural, physiological or behavioural in response to the environment. The mechanism of life processes, such as respiration, excretion, etc., vary from environment to environment. These responses occur within the framework of natural selection acting over evolutionary time.

2.8 DEATH

Nothing goes on forever, including the capacity of a normal and well-nourished cell to keep dividing. When a cell becomes senescent, it stops dividing but remains metabolically active for a time so that it gradually degenerates. We know that life is lost due to death and the organism reproduces to compensate for this loss of life. In biology, death has a great significance. All living plants and animals and the non living stones, rocks, etc. are formed of matter. They are cycled biologically. When organisms dic, microorganisms decompose the dead organisms. As a result, the elements of which the living bodies are constituted, such as C, H, N, O, Ca, K, P and S, are freed from their covalent bonds and return to the ecosystem. This is an example of recycling of elements between living and non-living matter. Such recycling actually maintains the balance of matter in nature.

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SUMMARY

Life is the property that distinguishes living from the non-living objects. Living organisms are highly organised and complex entities formed of one or more cells, carry out and control numerous chemical reactions. Biological organisation starts with chemical organisation of life, passes through organism level and ends in ecosystems and the biosphere. Atoms are the lowest unit at the molecular level, whereas the cells are the smallest unit at organismal level. Atoms combine to form molecules, which undergo chemical reactions to form organelles. Several organelles are contained in the cell. A group of cells meant for a specific function constitute tissue. In the organisms above the tissue level of organisation, many tissues become engaged to form organs, several organs constitute a system and there are many systems to take up the life process. Within the ecosystem, an individual forms the smallest unit. Individuals form population. Populations of different species living in the same area make up a biological community. The community interactions integrated with non-living (abiotic) features of the environment form an ecosystem.

Chemical reactions are changes in the atomic composition or structural organisation of the constituting atoms of substances. All chemical reactions involve changes in the relationship of electrons with one another by shifting of atoms from one molecule or ionic compound to another, Such shifting results in the formation or breaking of chemical bonds but does not produce any change in the number or identity of the atoms. Carbon is the principal structural element of a living cell. Though carbon dioxide contains carbon, it is generally considered inorganic and is the principal inorganic source of carbon. Before it can take part in a chemical reaction, CO_2 needs to get dissolved in water. Molecular oxygen (O_2) constitutes approximately 21per cent of the atmosphere. It is essential for life and is utilised by most plants and animals in the process of extracting energy from the nutrients. The green plants release oxygen during photosynthesis, which is the source of all atmospheric molecular oxygen. Organic molecules consisting of carbon and hydrogen are called hydro-carbons. Organic molecules may be small and simple. Large and complex assemblies of simple biological molecules are called macromolecules. The four important macromolecules are carbohydrates, lipids, proteins, and the nucleic acids.

All proteins are formed of subunits called amino acids. Only 20 different amino acids, which are made primarily of carbon, hydrogen, oxygen and nitrogen are required for proteins. Proteins make up the main structural and functional components of cells. They can also serve as biological catalysts or enzymes. Nucleic acids are long polymers of repeating subunits called nucleotides. The nuleotides are formed of a pentose sugar, a phosphate group and an inorganic nitrogenous base – purine or pyrimidine. The sugar is ribose in RNA and deoxyribose in DNA molecules. There are four types of nitrogenous bases, two of purines - adenine and guanine, and two of pyrimidines - cystosine and thymine.

Living systems have regulatory mechanisms for adjusting with the fluctuation of their external environment. Many organisms maintain a balanced (steady-state) internal environment by employing physiological and morphological or behavioural mechanisms. Majority of vertebrates and plants lose most of their thermal energy to their environments and are ectothermic and poikilothermous. Mammals, birds and a few fishes are endothermic and homeothermous because they retain thermal energy with the aid of thermoreceptors.

Growth occurs due to synthesis of protoplasmic and apoplasmic substances. Organisms grow by cell proliferation, cell enlargement and secretion. It is the result of a process called metabolism. Development involves growth, morphogenesis and differentiation. Reproduction involves the production of copies of the organisms and is necessary for the continuation of life and to compensate for the loss of life. Organisms reproduce by asexual or sexual means and pass their hereditary materials to their offspring. Adaptation is any characteristic of an organism, which makes it better suited to its environment. Adaptation may be structural, physiological or behavioural. In biology, death has a great significance. All living organisms and the non-living substances are formed of matter. Living organisms are the biotic and the non-livings are the abiotic features of the environment. When organisms die, microorganisms decompose the dead materials and the elements are freed, returned to the ecosystem and recycled. Such a recycling actually maintains the balance of matter in the nature.

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EXERCISES

1. What are the basic characteristics that unify all living organisms?
2. Give a schematic representation of the levels of biological organisation and the related branches of science.
3. Fill in the blanks
   (i) _____are the building blocks of matter and form the lowest biological unit.
   (ii) The three subatomic particles relevant for life are _____, _____ and _____.
   (iii) Each proton has _____ unit of positive charge and each electron has _____ unit of negative charge, whereas a neutron has _____ charge.
   (iv) An _____ is the smallest part of an element.
   (v) Trace elements constitute _____ percent of the organism's body weight and are of immense value as _____ nutrients of the organisms.
   (vi) The element iodine is necessary for the vertebrates as an ingredient of a hormone produced by the _____ gland.
   (vii) Humans need a daily intake of only _____ mg of iodine in the diet to protect them from the disease called _____.
   (viii) Atoms combine by chemical bonding and form _____.
   (ix) Sodium chloride is an important lattice of _____ in which the atoms are held together by _____bonds.
   (x) Sodium and Chloride ions participate in the _____of materials through cell membranes.
   (xi) A chemical reaction in which energy from outside source is needed for initiation of chemical reaction is called _____ reaction.
   (xii) A chemical reaction that yields energy and does not require energy from outside source is called _____ reaction.
   (xiii) Activation energy is the energy that is required to _____chemical bonds.

4. Why weak chemical bonds are advantageous for the chemistry of life? Explain your answer taking the common table salt as an example.
5. Explain feedback mechanism.
6. Describe the macromolecule that acts as the main energy source of cells.
7. Complete the following table.

Summary of the Major Classes of Macromolecules

Class	Chemical Elements	Monomer Subunits	Major Roles
1. _____	C, H, O, N, P	2. _____	3. (a) _____ (b) _____
4. _____	5. _____	6. _____	(a) Store energy (b) Form biomembranes (c) Hormones
Proteins	7. _____	8. _____	9. (a) _____ (b) _____
10. _____	11. _____	12. _____	13. _____

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8. How energy flows in the biological systems ?
9. What do you understand by 'free' energy ?
10. What is an open system? How does it differ from a closed system ?
11. What is metabolism? Mention the role of enzyme in metabolism.
12. Differentiate between anabolic and catabolic pathways. How are the two pathways regulated ?
13. Define homeostasis. Briefly describe the two broad mechanisms of temperature regulation in the different groups of vertebrates.
14. Explain the mechanism of feed back loop with special reference to homeothermy in humans.
15. What is growth ? How growth is related to metabolism ?
16. What are the three important characteristics of development? Discuss the interrelationship between growth, development and reproduction in the maintenance of life.
17. Define adaptation. Explain the phenomenon with a suitable example.
18. What is death? Mention the biological significance of death.

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Chapter 3

Origin and Evolution of Life

Life had a beginning. Since when life appeared on Earth? What is the mechanism involved in the origin of life ? Such questions are very difficult to answer because we cannot go back in time and observe life's beginning. Since its appearance, life on Earth has changed through time. Hence, the history of life actually comprises two events — first, the origin of life and, second, the mechanism involved in the changes of living organisms through time or evolution of life.

3.1 ORIGIN OF LIFE

The oldest surviving terrestrial rocks, about 4.3 billion years old, contain no definite trace of life, at least not recognisable as yet. Some rocks, about 3.9 billion years old, contain carbonates. Geologists interpret that these carbonates have resulted from life processes. Therefore, life was present on Earth about 3.9 billion years ago. However, the oldest microfossils discovered so far are that of photosynthetic cyanobacteria that appeared 3.3 to 3.5 billion years ago. Given this background, we can only speculate how did life originate and when.

Theories on the Origin of Life

Many theories have been advanced to explain the origin of life. Most of the ideas on the origin of life fall into one of four categories.

(i) Special Creation : This idea embodies that life on Earth is a special or divine creation of one or more superior, intelligent and all-powerful God. It also attributes the origin of life to a supernatural or vitalistic event at a particular time in the past. It upholds that life is immutable and has not changed ever since its origin.

(ii) Spontaneous Generation : According to this theory, life originated spontaneously from lifeless matter — **abiogenesis**. For example, frogs could arise from moist soil. However, this idea had been put to rest when Louis Pasteur (1862) provided proof that micro-organisms come only from other micro-organisms (refer Fig. 1.2). Since then, it is being widely accepted that life originates from pre-existing life — **biogenesis**.

(iii) Extraterrestrial or Cosmic Origin : This idea holds that life is coeternal with matter without any beginning. This notion advocates that life could have originated once or several times in various parts of the Galaxy in the Universe. The alternative name of this **cosmozoan theory** is **panspermia**, which holds that 'spores' or 'seeds' (sperm) of extraterrestrial origin might have infected the barren Earth at the time of its origin or afterwards. Likelihood of this notion is remote because the hazards known to exist in the interplanetary space are not conducive to life. These non-conducive conditions are extremely low temperatures, lack of atmosphere, utter dryness and very high flux of cosmic and ultraviolet radiation from the sun.

(iv) Terrestrial or Abiogenic Origin : This idea holds that life arose by a series of sequential chemical reactions. There is a general consensus of scientific opinions that life originated on Earth from collections of organic molecules that were produced early in the history of Earth.

Origin of Earth

Origin of life is inseparably linked with the origin of Earth. In fact, the history of life is a chronicle of a restless Earth. The ***Big Bang Theory*** proposes that the universe had an explosive beginning. Universe originated about 15 billion years ago by a big bang (thermonuclear explosion) of a dense entity (Fig 3.1). About 4.5 billion years ago, our solar system was probably created when the gaseous cloud called solar nebula

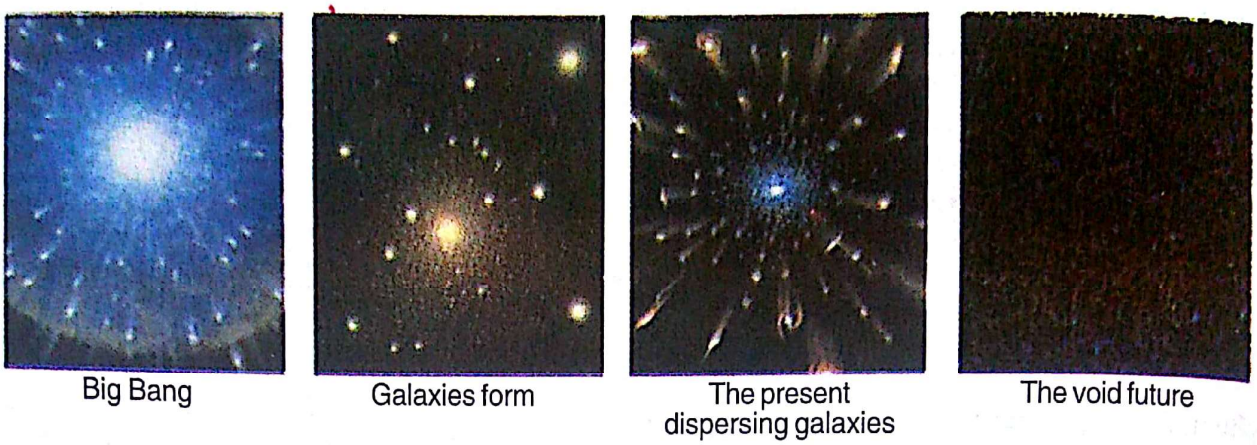

Fig. 3.1 *Schematic diagram of the Big Bang theory*

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started to collapse under the force of its own gravity until it became a flattened spinning disc of atoms and particles(Fig. 3.2). Its central region heated up and became a star, the Sun. Atoms and the dust grains orbiting the centre of the gaseous disc then aggregated into clumps (about the size of asteroids). These clumps finally swept up all particles and grew to the Planetary size, such as Mercury, Venus, Earth, Mars, Jupiter, Saturn, Uranus, Neptune and Pluto. These planets and the Asteroid belt between Mars and Jupiter to the solar system originated some 4.6 billion years ago. The Earth consisted of a solid phase, the lithosphere and a gaseous envelope surrounding it, the atmosphere. The liquid phase, the hydrosphere, appeared later when the Earth cooled down to a temperature below 100° C.

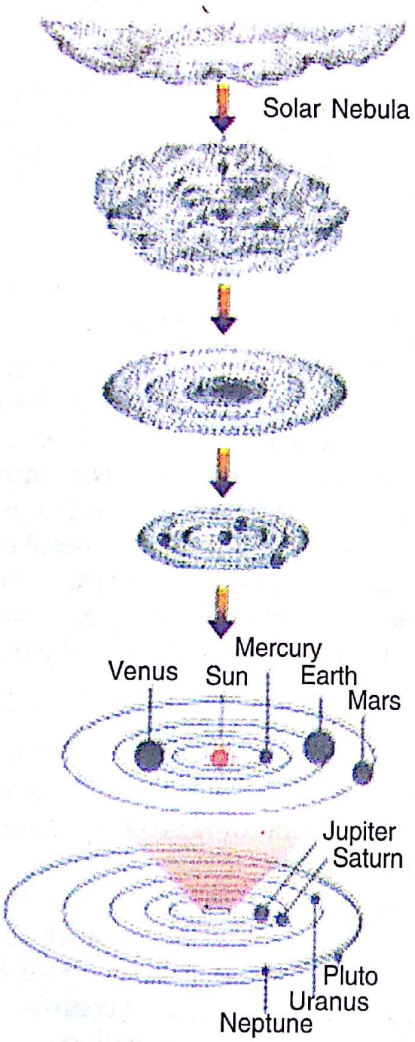

Fig. 3.2 *Schematic representation of condensation of solar nebula*

Abiogenic or Chemical Origin of Life

Majority of the scientists are of the opinion that life originated from inanimate matter. Since the theory of **abiogenic origin or chemical evolution** of life is the only one that provides an explanation, which can be tested, most scientists have tentatively accepted it. Consequently, we are to limit the scope of our inquiry to this scientific approach.

Oparin-Haldane Hypothesis

Alexander I. Oparin (1894-1980), a Russian biochemist, and J. B. S. Haldane (1892-1964), a British scientist, put forward the concept that the first living thing evolved from non-living things. They also suggested what the sequence of events might have been. In 1923, Oparin postulated that life originated on Earth at some point of time in the remote past and under the conditions no longer observed. In his book, *The Origin of Life* (1938), Oparin submitted that *abiogenesis first, but biogenesis ever since.* Oparin's theory is known as **primary abiogenesis.**

According to Oparin and Haldane (1929), **spontaneous generation** of early molecules might have taken place if the Earth once had more reducing atmosphere compared to the present oxidising atmosphere. Moreover, the transformation of those lifeless chemicals into living matter extended over some one billion years. Oparin and Haldane agreed that the primeval Earth contained little, if at all, oxygen. Perhaps, in the primitive atmosphere oxygen in the free gaseous state was virtually absent. However, oxygen remained bound in water and metallic oxides on the surface of rocks and its particles. Geological record shows the presence of such reduced materials like uranite, pyrite, etc. in the sediments, implying that the conditions then were strongly reducing (nonoxygenic). Therefore, no degradation of any organic compound arising in the primeval Earth could have taken place. As there was no ozone layer in the atmosphere any absorption of UV radiations that is lethal to our present lives was possible in the primordial Earth. The early gas cloud was rich in hydrogen, being present in the form of methane (CH_4), ammonia (NH_3) and water (H_2O). Thus, carbon, nitrogen and oxygen were available in the form of their saturated compounds. Moreover, the atmospheric water vapour condensed into drops of water and fell as rain that rolled down the rock surfaces and accumulated to form liquid pools and oceans. In the process, erosion of rocks and washing of minerals (e.g. chlorides and phosphates) into the oceans were inevitable. Thus, Haldane's **hot dilute soup** was produced and the stage was set for combination of various chemical elements.

Atmospheric chemicals and those in water produced small precursor molecules, like amino acids, sugars, nitrogenous bases etc. These precursor molecules then combined resulting in the appearance of proteins, polysaccharides and nucleic acids. The energy sources for such reactions of organic synthesis were the UV radiations (solar radiation), electrical discharges (lightning), intense dry heat (volcanic eruption) and radioactive decay of various elements on the Earth's surface. Once formed, the organic molecules accumulated in water because their degradation was extremely slow in the absence of any life or enzyme catalysts (Fig. 3.3). Such transformation is not possible in the present oxidising atmosphere because oxygen or micro-organisms will decompose or destroy the living particle that may arise by mere chance.

Experimental Evidence for Abiogenic Molecular Evolution of Life

Harold C. Urey (1893-1981), an Astronomer, accorded the first adequate recognition of Oparin-Haldane's view on the origin of life in 1952. Urey asked his student Stanely L. Miller, a biochemist, to replicate the primordial atmosphere as propounded by Oparin and Haldane. Miller (1953) made the first successful simulation experiment to assess the validity of the claim for origin of organic molecules in the primeval Earth condition (Fig. 3.4). Miller sealed in a spark chamber a mixture of water (H_2O), methane (CH_4), ammonia (NH_3) and hydrogen gas (H_2). He made arrangement for insertion of two electrodes to provide electrical energy (simulation of lightning) to the spark chamber. The spark chamber was connected to another flask with arrangement for boiling water (provision for evaporation). The other end of the spark chamber was connected to a trap by a tube that passed through a condenser (an arrangement for condensation and collection of aqueous solution, equivalent to rain and Haldane's soup). The trap, in turn, was connected with the flask for boiling water (arrangement for circulation). The control apparatus contained every arrangement except that it was devoid of energy source.

After eighteen days, significant amount of the simple major organic compounds (monomers), such as amino acids and peptide chains, began to appear in the aqueous sample of the experimental set. On the contrary, insignificant amount of organic molecules was formed in the control apparatus. Therefore, the obvious inference was that abiotic synthesis of organic monomers occurred in the simulated experimental condition. By analogy, such synthesis could have occurred in the primitive atmospheric condition. Later on many scientists repeated Miller's experiment using slightly different starting materials and UV radiation or other energy sources. All of them could successfully synthesise amino acid and related compounds. With hydrogen cyanide (HCN) even adenine and other nitrogen bases were produced.

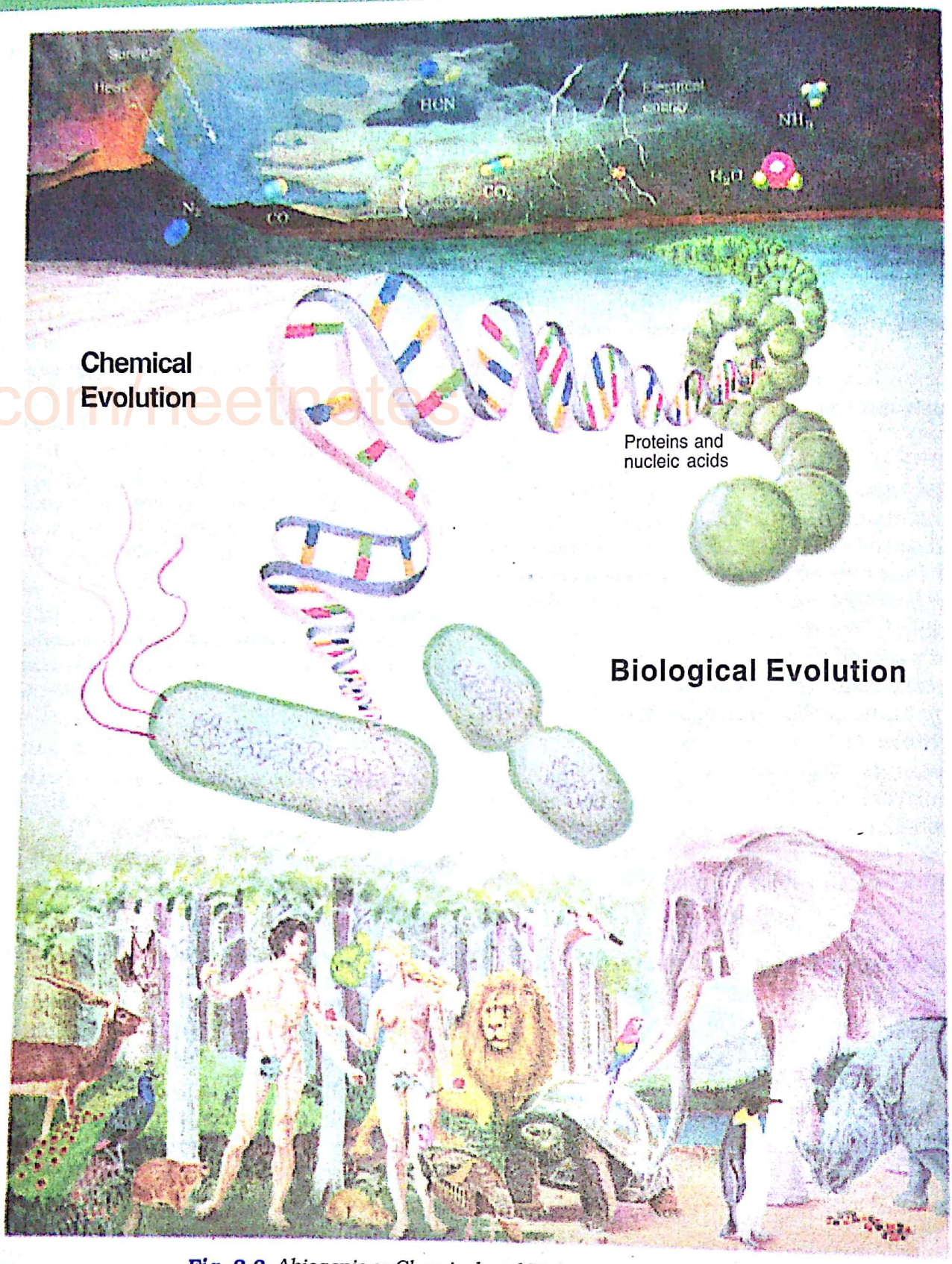

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Fig. 3.3 *Abiogenic or Chemical and Biological evolution of life*

Polymers and the Problem of Condensation

Such abiogenically produced building blocks of life as amino acids, nitrogenous bases, oxides of copper, iron and saturated hydrides, etc. were in low concentrations because they reached a steady state. Consequently, 'the hot soup' remained always dilute and this form placed a limit on chemical complexity. We know that the important biomolecules, such as polysaccharides (carbohydrates), polypeptides (proteins),

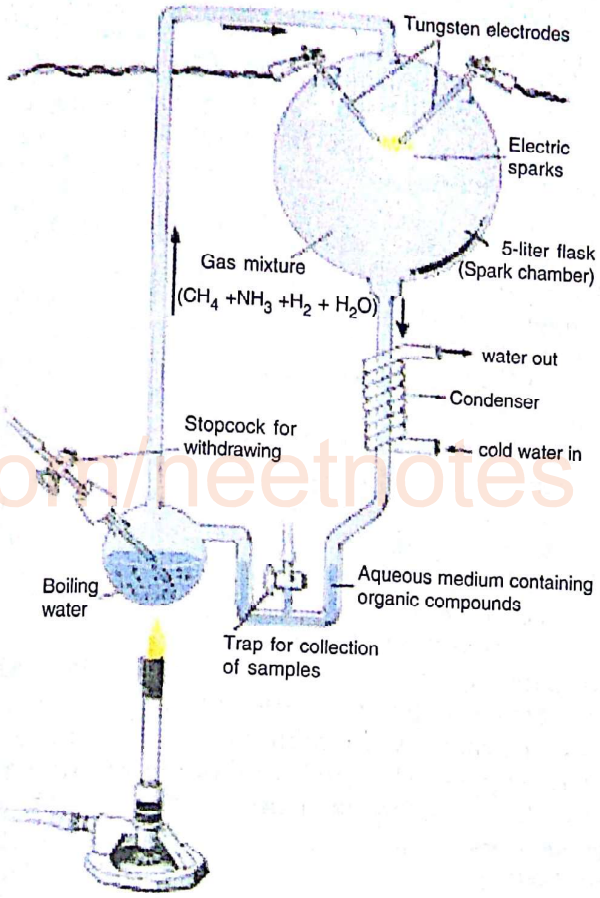

Fig. 3.4 *Miller's experiment to prove the origin of life in the simulated primeval atmosphere*

polynucleotides (nucleic acids), are complex polymers. Formation of compounds of greater complexity (polymeric biomolecules) from compounds of modest organic monomers in the primordial Earth, therefore, posed a problem in the success of molecular evolution of life. A mechanism to concentrate monomers and thus favouring polymerisation was necessary as evolutionary breakthrough.

The major requirement for promoting polymerisation is the availability of adequate sources of energy and removal (evaporation) of water from the surface of the reactants so that they can concentrate and prevent depolymerisation. In the primeval atmosphere electrical discharge, lightning, solar energy, ATP and polyphosphates might have provided the sources of energy and evaporation of water resulted from solar heat. Thus, polymers that formed faster or were more stable would have come to predominate. High concentrations of polymers, in turn, would have stimulated further polymerisation by shifting chemical equilibrium from unstable monomers to stable polymers.

Enclosing the Prebiotic Systems

The experiments of Miller and other scientists demonstrate that prebiotic molecules could have been formed under the conditions which most likely existed on early Earth. Still, the formation of prebiotic soup of small molecules does not necessarily lead to the origin of life. For origin of life, atleast three conditions needed to have been fulfilled:

(i) There must have been a supply of replicators i.e. self-producing molecules.
(ii) Copying of these replicators must have been subject to error via mutation.
(iii) The system of replicators must have required a perpetual supply of free energy and partial isolation from the general environment.

The high temperature prevailing in early Earth would have easily fulfilled the second condition, that is, the requirement of mutation. The thermal motion would have continually altered the prebiotic molecules.

The third condition, partial isolation, has been attained within aggregates of artificially produced prebiotic molecules. These aggregates called **protobionts** can separate combinations of molecules from the surroundings; maintain an internal environment but are unable to reproduce. Two important protobionts are **coacervates** and **microspheres**. Oparin (1924) observed that if a mixture of a large protein and a polysaccharide is shaken, coacervates form. Their interiors, which are primarily protein and polysaccharide, with some water, become separated from the surrounding aqueous solution. The later has much lower concentration of proteins and polysaccharide. Oparin's coacervates also exhibit a simple form of metabolism. As these coacervates do not have lipid outer membranes and cannot reproduce, they fail to fulfil the requirement as a candidate of probable precursors of life.

Microspheres form when mixtures of artificially produced organic compounds are mixed with cool water. If the mixture contains lipids, the surface of the microspheres consists of a lipid bilayer, reminiscent to the lipid bilayer of cell membranes. Sydney Fox (1950) obtained

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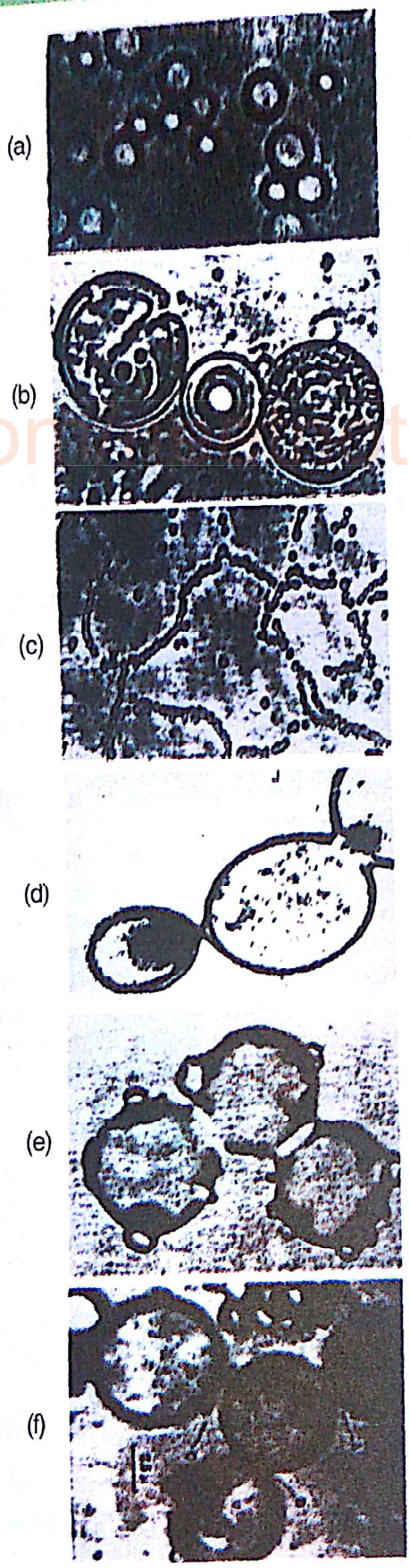

Fig. 3.5 *Forms of protenoid microspheres (a-f)*

protenoid microspheres (Fig. 3.5) by heating a mixture of dry amino acids between 130 to 180° C and later cooling them in water. They were about 1 to 2 μm in diameter, similar to the size and shape of coccoid bacteria. Further, they could be induced to constrict in a process that superficially resembles budding in bacteria and fungi. Their main drawback is that they have limited diversity. Thus the mechanism of partial isolation leading to the origin of protobionts still remains a subject of speculation.

When light shines on protobionts that have chromophores (molecules that absorb visible light) in their membranes, electrical potentials develop across the membrane. Such protobionts become energy transducing devices. Given the fact that the principal route of biological energy flow is from solar radiation to an oxidised and reduced compound, it may be presumed that chromophores were most likely present in the lipid membranes of some protobionts. As such, the earliest life (protolife) might have been driven primarily by solar energy. The nucleic acids appear to be good candidates for fulfilling the first condition, that is, the supply of replicators in the prebiotic life. The experiments of Miller yielded purine and pyrimidine constituents of nucleic acids. We know that the enzymes that control the types and rate of reactions within organisms are proteins. Proteins are synthesised by a process that begins with the transcription of information from DNA to messenger RNA (mRNA). This system of protein synthesis, **DNA → RNA → Protein**, probably evolved gradually from much simpler processes.

How could such a system have evolved if proteins needed nucleic acids to form and nucleic acids needed proteins to catalyse their replication? Could protcins have evolved before nucleic acids? Current data showing that some ribosomal RNA (rRNA) have catalytic as well as informational properties may help to resolve the chicken-and-egg paradox of metabolism first or genes first. The first genetic code was based on RNA that catalysed its own replication as well as catalysing other chemical reactions. The accumulated products of RNA-catalysed reactions could then participate in other reactions and form structures. For example, RNAs could have catalysed the formation of lipid-like molecules that could have, in turn, formed

plasma membrane and proteins. Further, this protein could have been successful to catalyse the synthesis of other proteins. Eventually, the proteins might have taken over most enzymatic functions because they are better catalysts than RNA and are capable of more specific activities. If the first cells used RNA as their hereditary molecule, then DNA evolved from an RNA template. DNA probably did not evolve as a hereditary molecule until RNA-based life became enclosed in membranes. Once cells evolved, DNA probably replaced RNA as the genetic materials for most organisms.

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The Earliest Cells

The first living organisms, having arisen in a sea of organic molecules and in contact with an atmosphere, lacking oxygen (reducing atmosphere), presumably obtained energy by the fermentation of some of these organic molecules. The earliest organisms, presumably, were anaerobes, capable of respiration in the absence of oxygen. They must have depended on the existing organic molecules for their nutrition and, thus, heterotrophs. However, long before the supply of existing organic molecules was exhausted, some of the heterotrophs might have evolved into autotrophs. These organisms were capable of producing their own organic molecules by chemosynthesis or photosynthesis. One of the by-products of photosynthesis is gaseous oxygen. The emergence of photosynthesis was a turning point because this process changed the atmosphere of the Earth and, hence, initiated evolution of life toward its diversities.

3.2 THE MEANING OF EVOLUTION

Observation unveils that the diverse types of animals bear some common characters. For example, amphibians, reptiles and mammals have limbs for locomotion on land, fishes have fins for swimming in water, birds have wings for flying. A close scrutiny reveals that the limbs, fins and wings are formed on the same basic structural plan. All such examples can be explained if we consider that the diverse groups of organisms share a common ancestor from whom they have diverged and formed two different species. With the passage of time a single ancestral lineage (an evolutionary sequence, arranged in linear order from an ancestral group to a descendant group) has produced two or more lineages that also diverged over time. Such process of change in biological system is called evolution.

The word **evolution** (L. *evolvere*) means 'to unfold or unroll' or to reveal hidden potentialities. In its broadest sense, evolution simply means an orderly 'change' from one condition to another. For instance, the planets and stars change in between their birth and death. This is stellar evolution. Matter changes in time. This is inorganic evolution. The changes in the properties of population of organisms or groups of such populations over the course of generations are considered as biological or organic evolution. It is a process of cumulative change of living populations and in the descendant populations of organisms. In other words, it is descent with modifications. In general, the diversities of life include both the differences and similarities, and the characteristics of organisms, both adaptive and non-adaptive. These are the great themes of evolutionary biology. According to Theodosius Dobzhansky (1973), *nothing in biology makes sense except in the light of evolution.*

3.3 IDEAS OF EVOLUTION BEFORE DARWIN

Evolutionary biology has a much longer and richer history than most of us realise. The contributions of ancient people are often slighted. All theories of evolution, from Darwin to the present day, are permeated with numerous earlier concepts and theories.

Ancient Indian Thoughts on Evolution

The ancient Indians succeeded in understanding the origin and evolution of life in its broad outline. The ancient Indian texts of philosophy and Ayurveda deal with the origin of life. Manu's texts in Sanskrit, *Manu Samhita* or *Manu-smriti* (about 200 AD) mentions about evolution.

Ancient Greek Thoughts on Evolution

The first theories of evolution came about 2000 years before Darwin.

According to Plato (428-348 BC) each species was an unchanging Ideal Form *(eidos)*. All earthly representatives are imperfect imitations of such true essence of

an ideal unseen world. Since, God is perfect everything that existed on Earth was His 'ideas'. Aristotle (384-322 BC) expanded Plato's idealistic concepts to a chainlike series of forms – each form representing a link in the progression from most imperfect to the most perfect. He called this ***Ladder of Nature*** or ***Scala Naturae*** (Fig. 3.6). This is also known as the ***Great Chain of Being***.

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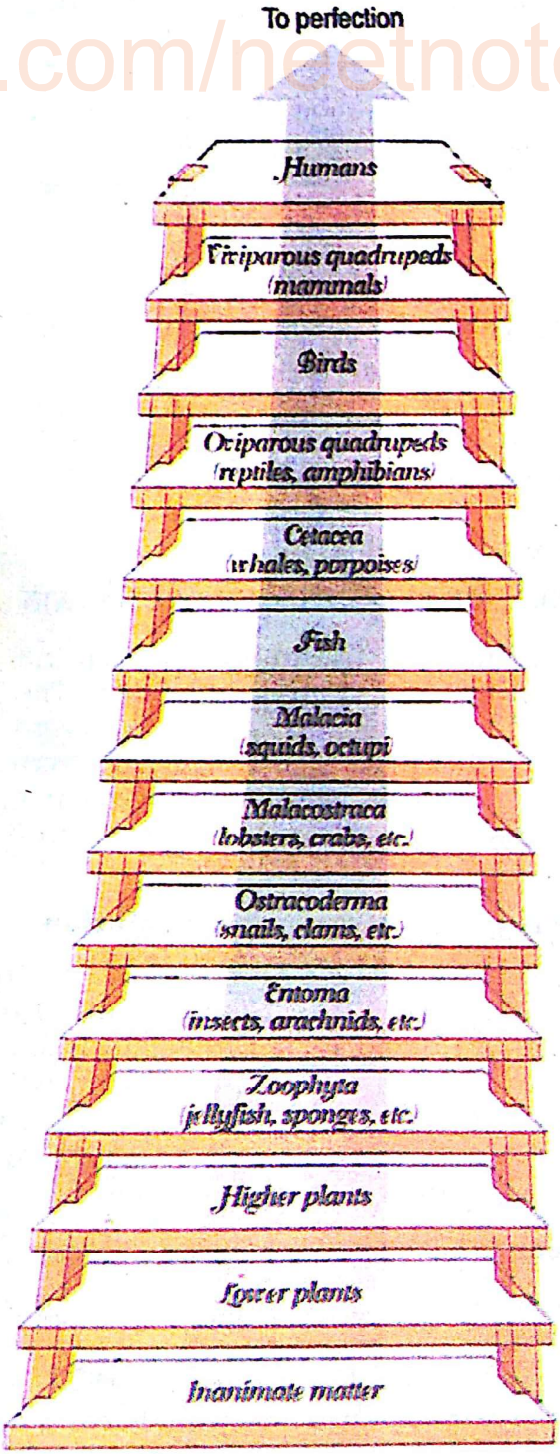

Fig. 3.6 *Aristotle's Ladder of Nature*

3.4 EVIDENCE OF EVOLUTION

Charles Robert Darwin (1809-1882), an English naturalist was the first to resolve all the previous perplexities of evolution. Several different lines of evidence convinced Darwin and his contemporaries that modern organisms arose by evolution from more ancient forms. Darwin documented evidence of evolution mainly on the basis of geographical distribution of species and fossil record. Biological evolution has left marks in the fossil record and in the historical vestiges evident in modern life. However, with the advancement of biology, new discoveries, including the revelations of molecular biology, continue to validate the evolutionary view of life.

Evidence from Biogeography

Biogeography means the study of geographical distribution of species. The story of Darwin's **theory of natural selection** begins in 1831, when he joined as an unpaid naturalist on a five year navigational mapping expedition of the British Admiralry aboard the ship H. M. S. Beagle.

During his voyage (Fig. 3.7), Darwin observed and studied a wide variety of plants and animals on continents and islands and in distant seas. Darwin spent a month in the black and lava-ridden islands of volcanic origin, the Galápagos Islands, which lie on the equator about 900 kilometre off the West Coast of South America. There, he found giant tortoises (*Sp.*; *galápago*), metre-long marine and land iguanas, many unusual plants, insects, lizards and sea shells.

The Galápagos Islands consisted of twenty-two different islands that were only a few miles apart. Darwin noticed that the Galápagos Islands have many endemic (native, found nowhere else) species of plants and animals. He was amazed to record that in the islands insect-eating warblers and woodpeckers were absent. Instead, various types of finches, a group of small black birds, which were originally seed-eating but have assumed insect-eating pattern, were present in the islands. These finches are often referred to as **Darwin's finches** (Fig. 3.8). He also observed that different geographical localities have similar habitats but house different species. The finches of different islands varied yet they were closely related to each other.

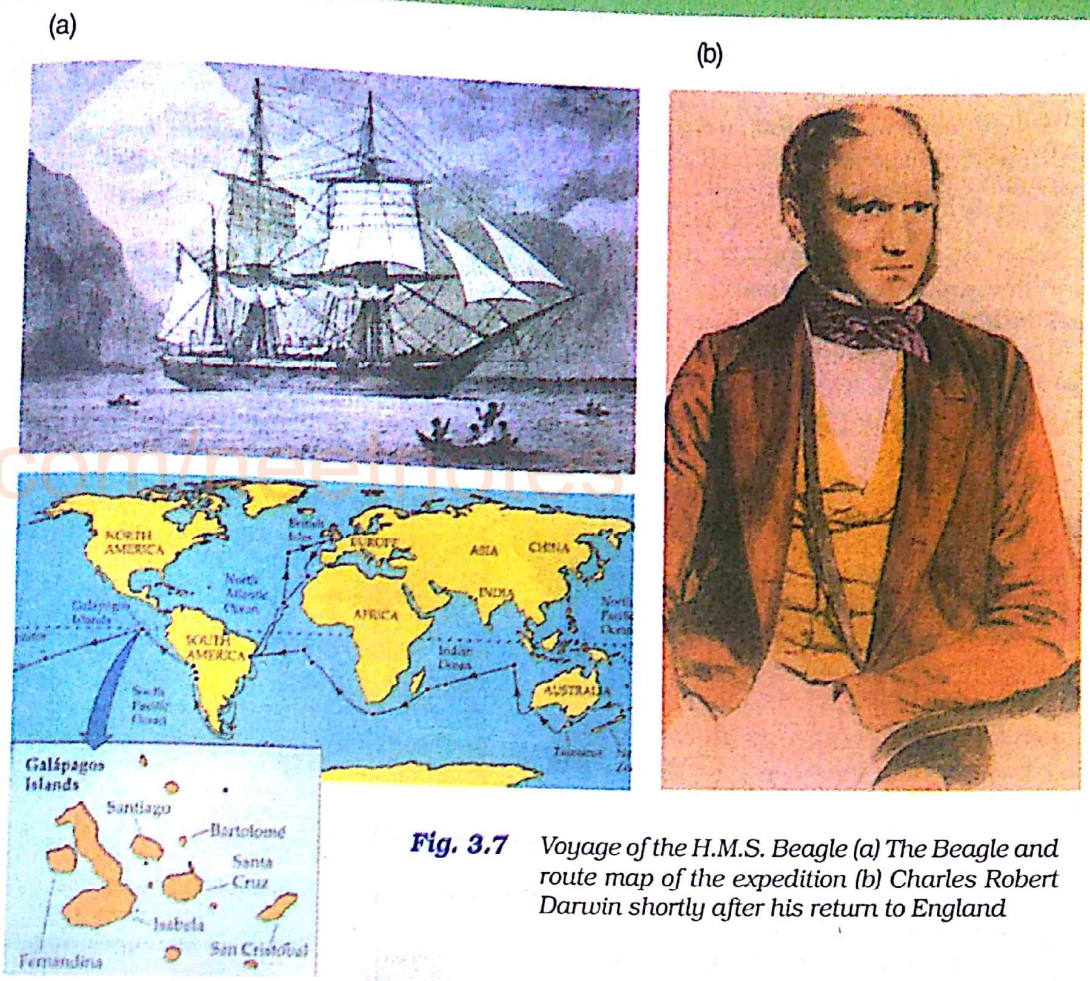

Fig. 3.7 *Voyage of the H.M.S. Beagle (a) The Beagle and route map of the expedition (b) Charles Robert Darwin shortly after his return to England*

Why the plants and animal species of the islands are closely related to species of the nearest mainland or neighbouring island ? Why the islands contain more different species of finches than the entire South American continent ? Darwin realised that such questions could be explained on the ground that from an ancestral group. Descendant populations could radiate into other areas, where the new environmental conditions brought about the suitable adaptation by evolution. He reasoned that after originating from a common ancestral seed-eating stock the finches radiated to different geographical areas and underwent profound adaptive changes, especially in the patterns of beak. Living in isolation for long period of time new kinds of finches emerged that could function and survive in the new habitats. Such an evolutionary process, giving rise to new species adapted to new habitats and ways of life, is called **adaptive radiation**.

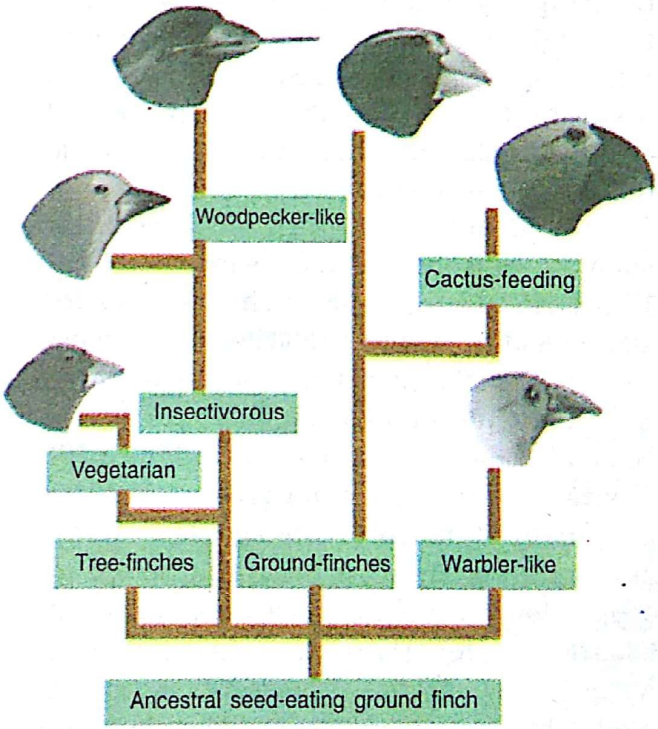

Fig. 3.8 *Darwin's Finches*

Evidence from Anatomy

The anatomical similarities among the species belonging to the same taxonomic category provide evidence for descent with modification.

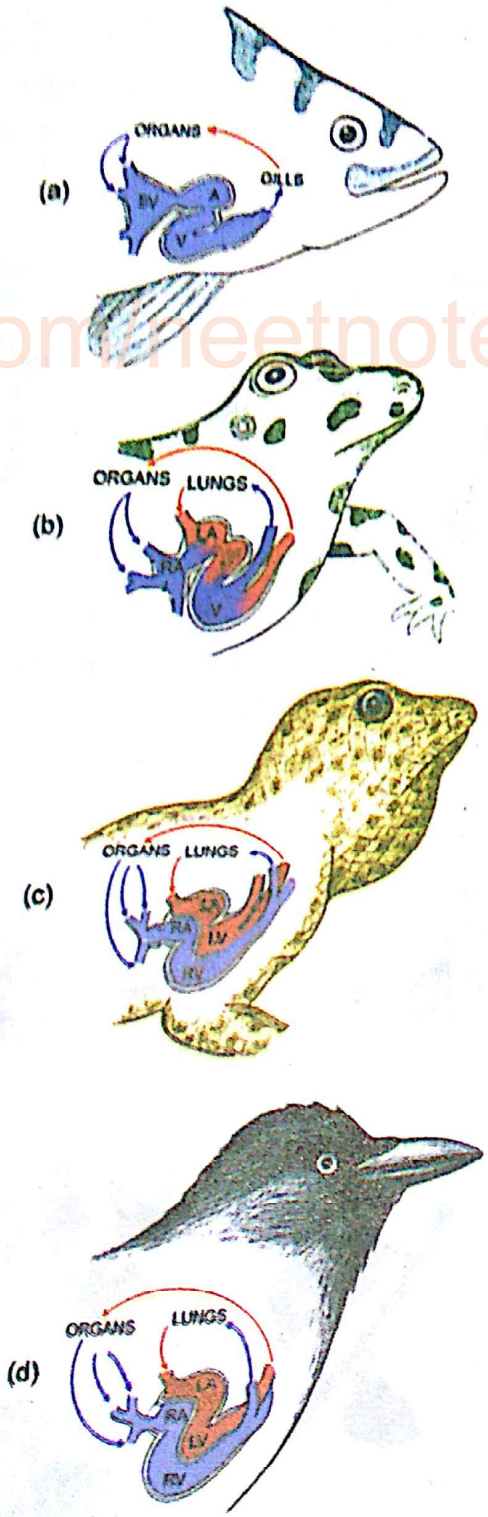

Fig. 3.11 *Homolgy and evolution of vertebrate heart (a) Fish (b) Frog (c) Lizard (d) Bird LA= Left auricle; RA = Right auricle; LV = Left ventricle; RV = Right ventricle; SV = sinus venosus*

Studies on the comparative anatomy of the heart of vertebrates indicate that the heart in all the groups has a basic structural plan. Such observations on basic similarities make sense if we accept that all mammals descended from a common ancestor. Richard Owen (1804-1892) introduced the term **homologous** to mean the organs of different species that are related to each other through common descent, although now functionally different.

A critical analysis of the anatomical information points out that the heart of vertebrates has changed in the course of evolution of mammals from fishes (Fig.3.11).

To elaborate, in fishes the heart is two-chambered, with one auricle and one ventricle. With the transition of fishes into land vertebrates (amphibians) and change in the mode of respiration, separation of oxygenated and deoxygenated blood among the terrestrial animals has become essential. As a result, the auricle has become divided into two compartments; it is two-chambered in all of the land animals. In order to achieve more perfection the ventricle shows a tendency of division. It is incompletely divided in reptiles, except the crocodiles. Further perfection in the separation of the two types of blood has been attained in the crocodiles, birds and mammals where the ventricle is completely divided. The heart is four-chambered. Similarly, their homologous structures shared in the brain by different vertebrates (Fig.3.12) are the cerebellum, cerebrum, medulla, olfactory bulbs and optic lobes .

Taking the endoskeleton, one can observe that the forearms of human, the wings of bat, the flippers of whales and the forelimbs of other vertebrates all are formed of the same basic skeletal elements. Also, they have a common structural plan. All of them contain bones named as humerus, radius, ulna, carpals, metacarpals and phalanges (Fig. 3.13a). Even more, all the bones are derived from the same part of the body.

In plants, a thorn of *Bougainvillea* differs from a tendril of *Cucurbita* in its function, but both are located in a similar (axillary) position. Thorns and tendrils are considered homologous (Fig 3.13b).

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Fig. 3.12 *Homologous structures: vertebrate brain (a) fish (b) frog (c) bird (d) cat (e) human being*

The converse of homologous organs is **analogous** organs. These are similar in function but are anatomically different and unrelated. For example, the wings of birds and the wings of butterfly, both of which are used for flying, are completely different in their anatomical framework. Neither do they have similar origin nor they have evolved from the same organ in a common ancestor. Even the strikingly similar analogous organs, such as the eye of an octopus and the eye of a mammal, differ in their retinal position. Similarly, the flippers of penguin (bird) and dolphin (mammal), which perform similar functions in these aquatic animals, have originated from different structures of two different lineages. Actually, the analogous organs are the result of convergent or parallel evolution in separate lineage. You might know about the existence of different modified roots, stems and leaves. Sweet potato is an underground tuberous root and potato is an underground stem but both the modifications are meant for storage of food. These are, therefore, examples of analogous organs.

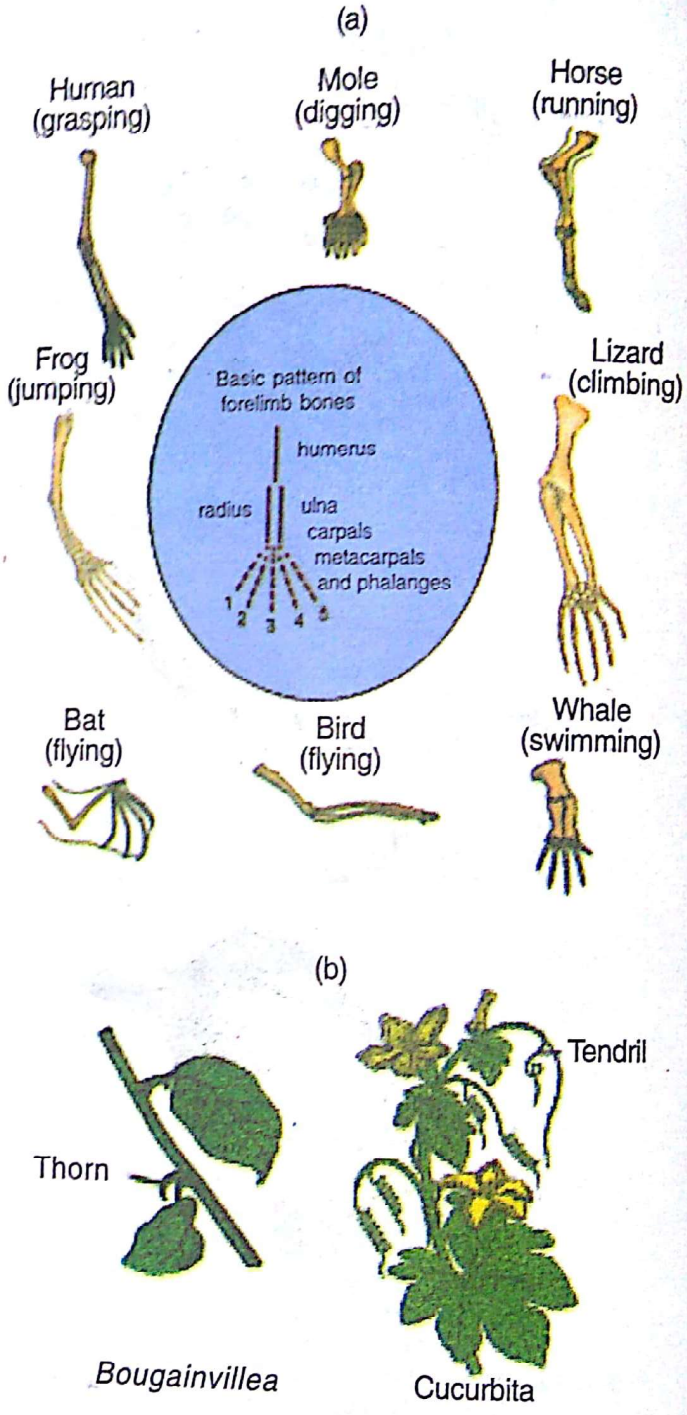

Fig. 3.13 *Homologous structures : (a) vertebrate forelimb, (b) thorns and tendrils in plants*

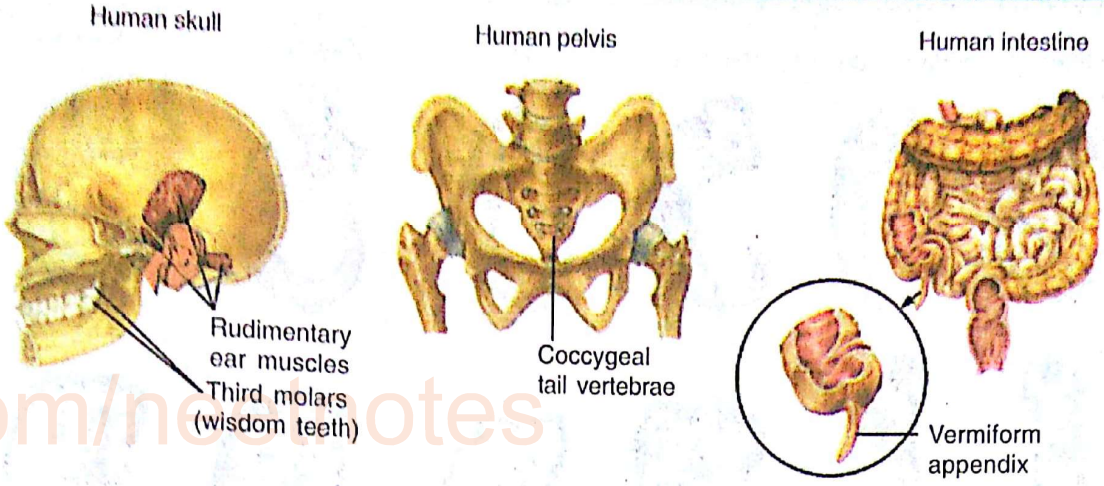

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Fig. 3.14 *Some vestigial organs*

Anatomical studies also help us to identify the structures that have lost some or all of the functions they used to perform earlier in their ancestors. Besides these studies also offer an evolutionary explanation of such rudimentary vestiges by stating that adaptations to new environment of the organism have made these structures redundant. Such structures are called **vestigial** organs. The rudiment of the reptilian jaw apparatus, the rudiment of the hind limbs of python and Greenland whales are some of the examples of vestigial organs. In humans, many vestigial structures (Fig. 3.14) indicate a relationship to other mammals, including the primates. For instance, muscles of the external ear and scalp are rudimentary and often non-functional. But these are common to many mammals where they are functional. The reduced tailbones and nictitating membrane of the eye, the appendix of the caecum, rudimentary body hair and wisdom teeth – all are examples of vestigial organs.

Evidence from Embryology

Embryos of the vertebrate series exhibit many features that are absent in the adults. To take an example, all the embryos of vertebrates, including humans, develop a row of vestigial gill slits just behind the head. Given the fact that gill slits are functional in the fishes only and not in the groups of vertebrates living on land, why the structure appears in the early embryonic stage of the land vertebrates? Answer can be found only on the premises that land vertebrates descended from the fishes that contained gill slits as an adaptation to aquatic respiration. However, the aquatic mammals (e.g. dolphins, whales, seals, porpoises, etc.) do not contain gill slits because their adaptation to aquatic life is secondary.

It was von Baer (1792-1867) who observed that the features such as brain, spinal cord, axial skeleton, aortic arches, etc., are common to all vertebrates. There are stages in development of embryos that vertebrates share (Fig. 3.15). Organisms that share common descent show underlying embryological patterns on which they build later their adult patterns. The special features, like hair (found in mammals only), feather (found in birds only), limbs (found in quadrupeds only), distinguish the various members of the group.

Ernst Haeckel (1834-1919) reinterpreted Baer's Law in 1868 in the light of evolution. This law held that ontogeny (development of the embryo) is recapitulation of phylogeny (the ancestral sequence). He viewed adult organisms as the embryonic stages of more advanced organisms. This view was summarised by his *Biogenetic Law: Ontogeny Recapitulates Phylogeny*. Interestingly, von Baer had disproved the biogenetic law before Haeckel stated it. By observing development, he noted that embryos never pass through the adult stages of other animals.

However, recapitulation has some value, although it is not a general phenomenon. For example, the seedlings of *Acacia* tree initially develop simple leaves which later transform into compound leaves. Another example is of the modern day oaks of southern United States.

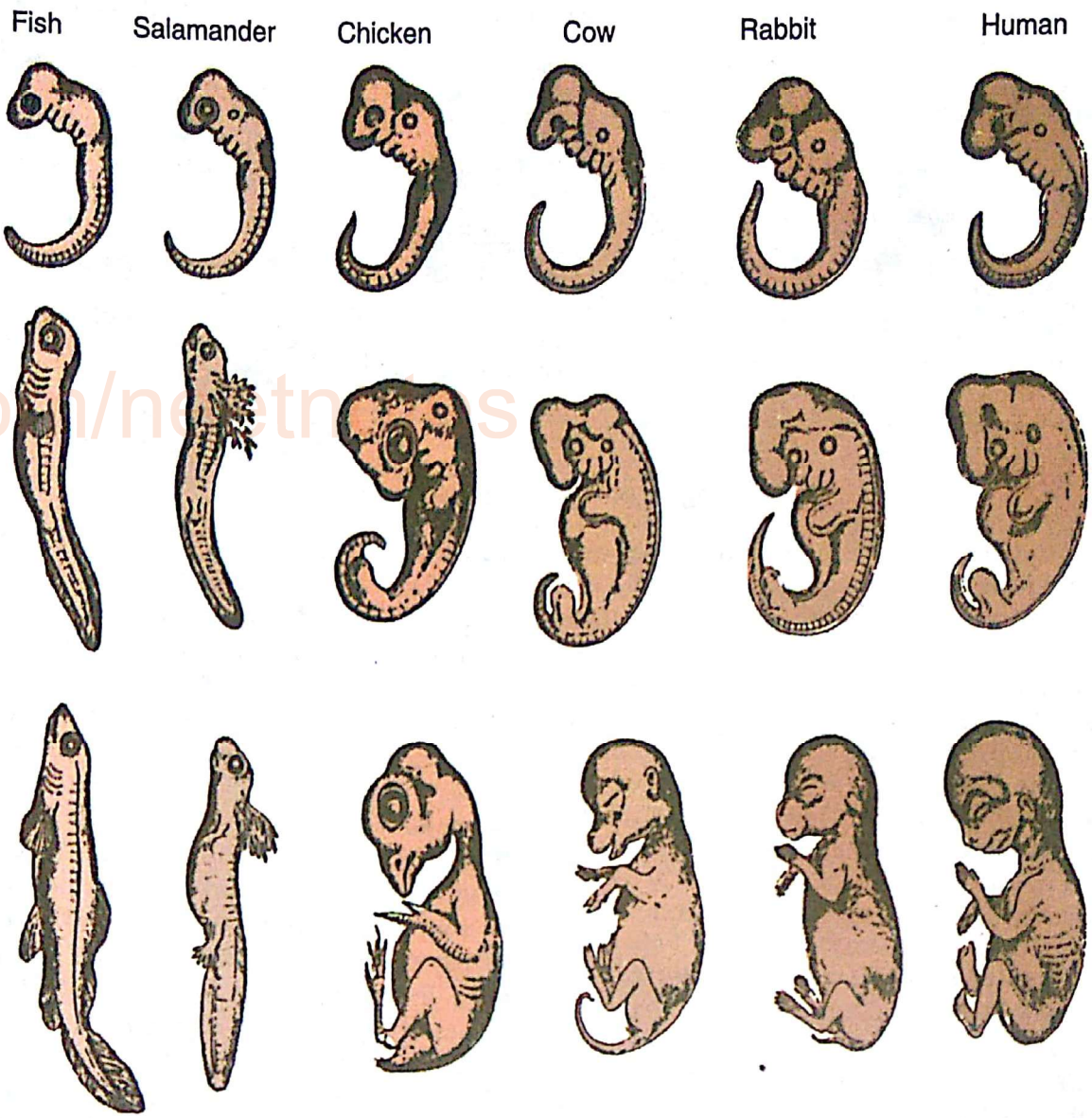

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Fig. 3.15 *Embryological evidence of evolution*

They retain their foliage throughout the year whereas the oaks of northern United States are deciduous and shed their leaves during winter. The southern species, on the basis of this character of leaves is considered to be more primitive than the northern oaks. However, the saplings of northern species are generally seen to retain their leaves during winter.

Evidence from the Fossil Record

Fossils are the preserved remains, tracks or traces of organisms that lived in the past. A body or body part may be preserved when organisms become buried in an acid bog, or under a layer of mud that cuts off oxygen, conditions that prevent decay. Sometimes, an impression of body surface (such as footprint), organic molecules (like oil) or even a coprolite (preserved excrement) may be preserved as fossil. Fossils are also created in sediment, the calcium in bone or other hard tissue mineralises and the surrounding sediment eventually forms rock. The fossils contained in layers of sedimentary rock reveal a history of life on Earth.

By dating the rocks in which the fossils occur, an accurate idea about the age of the fossils can be obtained. Rocks in deeper strata are generally older compared to those of the upper strata. Knowing the relative positions of the rocks and the rates of their erosion in different environments, the geologists of the nineteenth century determined the relative ages of

sedimentary rocks and their fossils. Assigning relative dates to fossils made it clear that some groups of species are older compared to others. Today, rocks are dated by measuring the degree of spontaneous decay of certain radioactive isotopes, which were locked within the rock when it was formed. This method is called absolute dating. Radioactive isotopes decay with a characteristic half-life (the time required to decay half the amount of radioactive isotopes). It is now possible to determine the actual age of rocks and the fossils in them. The **Geological Time Scale** (Table 3.1), thus prepared, consists of four major Eras; each era is subdivided into some Periods that refer to different stages of evolution of life on Earth.

When fossils are arrayed according to their age, from oldest to youngest often we get evidence for successive changes of the organism through ages. Today, the fossil records are far more complete than Darwin's time, particularly among the vertebrates. Different classes of vertebrate appear chronologically in the fossil records. Fossil fishes predate all other vertebrates, with

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Table 3.1 Evolution of Vertebrates (Hypothetical)

THE GEOLOGICAL TIME SCALE

ERA	PERIOD	EPOCH	AGE (MILLIONS OF YEARS)	SOME IMPORTANT EVENTS IN THE HISTORY OF LIFE
Cenozoic	Quaternary	Recent	0.01	Historic time
		Pleistocene	1.8	Ice ages; humans appear
	Tertiary	Pliocene	5	Ape like ancestors of humans appear
		Miocene	23	Continued radiation of mammals and angiosperms
		Oligocene	34	Origins of most modern mammalian orders, including apes
		Eocene	57	Angiosperm dominance increases; further increase in mammalian diversity
		Paleocene	65	Major radiation of mammals, birds, and pollinating insects
Mesozoic	Cretaceous		144	Flowering plants (angiosperms) appear; dinosaurs and many groups of organisms become extinct
	Jurassic		208	Gymnosperms continue as dominant plants; dinosaurs dominant; first birds
	Triassic		245	Gymnosperms dominate landscape; first dinosaurs and mammals
Paleozoic	Permian		285	Radiation of reptiles, origin of mammal-like reptiles and most modern orders of insects; extinction of many marine invertebrates
	Carboniferous		360	Extensive forests of vascular plants; first seed plants; origin of reptiles; amphibians dominant
	Devonian		408	Diversification of bony fishes; first amphibians dominant
	Silurian		438	Diversity of jawless vertebrates; colonization of land by plants and arthropods; origin of vascular plants
	Ordovician		505	First vertebrates (jawless fishes); marine algae abundant
	Cambrian		544	Origin of most invertebrate phyla; diverse algae
Precambrian			700	Origin of first animals
			1500	Oldest eukaryotic fossils
			250	Oxygen begins accumulating in atmosphere
			3500	Oldest definite fossils known (prokaryotes)
			4600	Approximate origin of Earth

amphibians next, followed by reptiles, then mammals and birds. Fossils have been found linking all major groups of vertebrates. Palaeontologists have discovered many transitional forms that link even older fossils to modern species. For example, a series of fossils documents the changes in skull shape and size that occurred as mammals evolved from reptiles.

The patterns of evolution of vertebrates (Fig. 3.16) and major groups of plants (Fig. 3.17) are conspicuously different.

The major groups of vascular plants have left relatively small number of fossils, which even show gaps. There are relatively few major lineages, and all the lineages are very distinct from one another. Instead of showing gradual and continuous change through time, the major lineages appear suddenly in the fossil record. After that they persisted with little fundamental change for hundreds of millions of years. The existence of many of the major subdivisions of the vascular plants living today can be recognised about 345 million years ago on the basis of their distinctive reproductive structure. All primitive land plants reproduce via tiny spores contained in the sporangia. The major taxonomic groups are distinguished by the position of sporangia on the plant.

The sporangia are terminal, located at the tip of the plant in the most primitive Psilopsida. These are placed at the base of the leaves in the Lycopsida (represented in the modern flora by *Lycopodium* and *Selaginella*). The sporangia are arranged in whorls at the top of the plant in Sphenopsida (horsetails). Fossil evidences document that these basic patterns have been

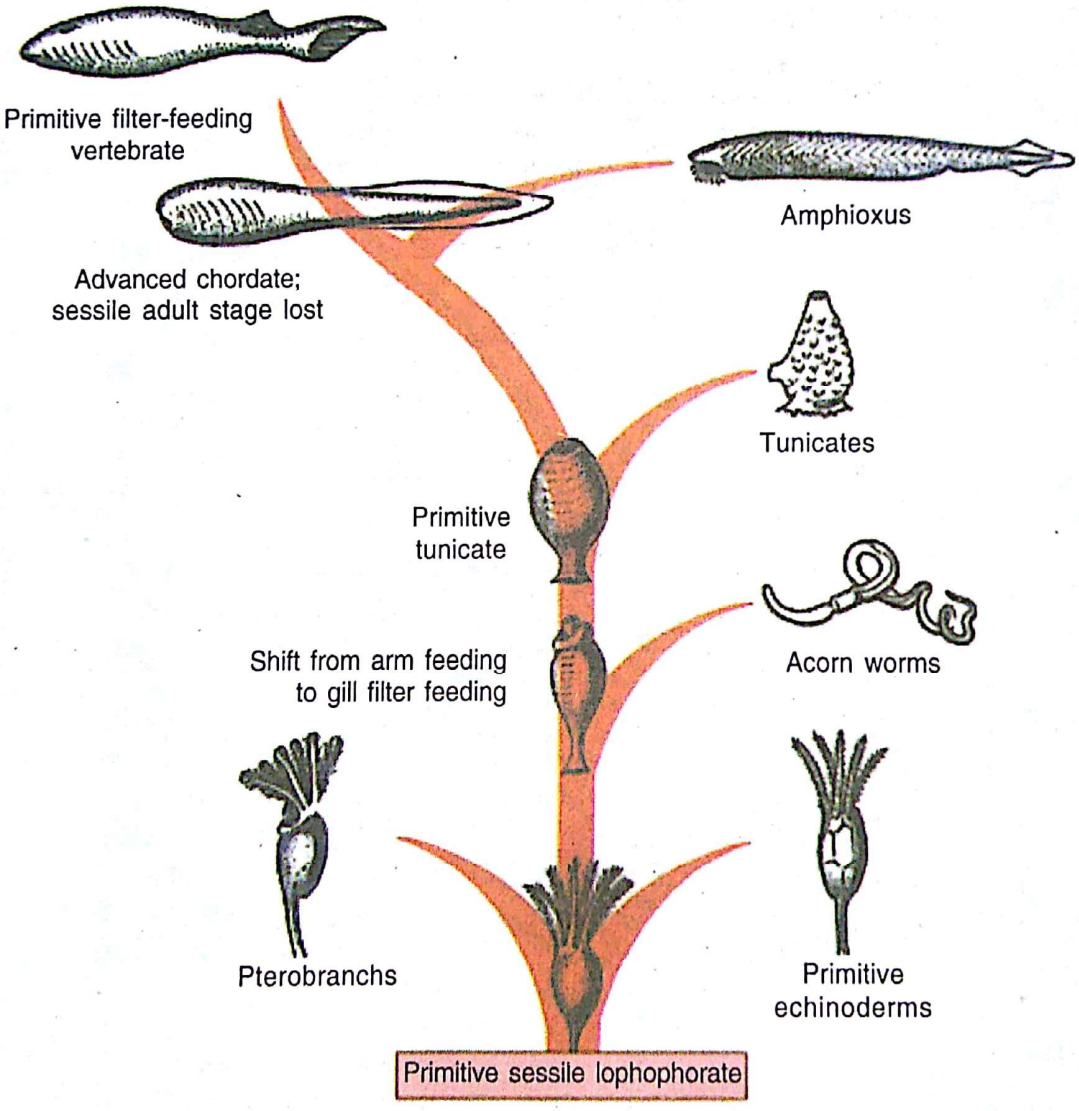

Fig. 3.16 *Evolution of vertebrates (Hypothetical)*

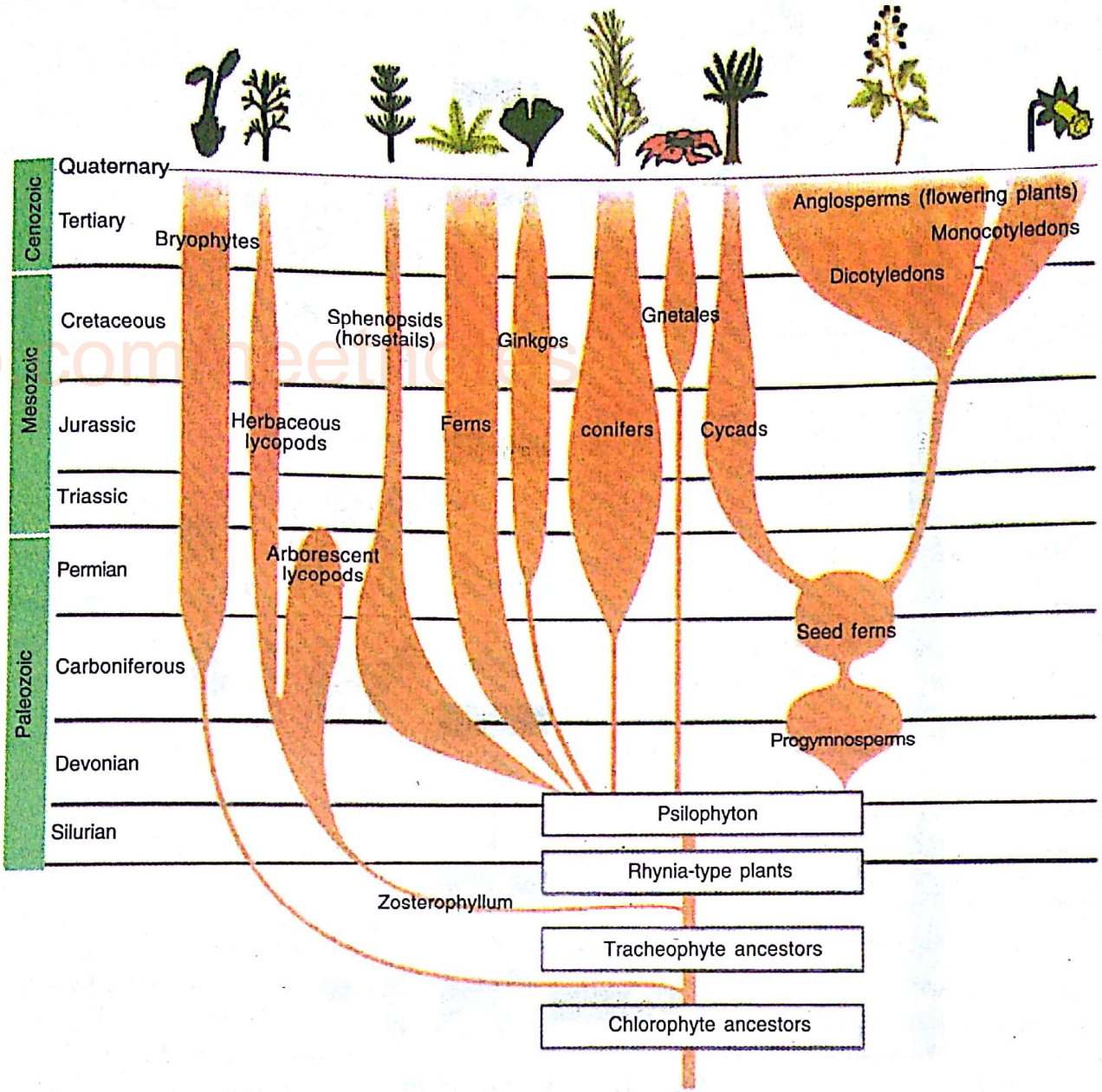

Fig. 3.17 *Evolution of plants (Hypothetical)*

maintained for more than 350 million years. Few, if any, intermediates are known between these patterns. The origin of seeds in the land plants was achieved about 345 million years ago in lineages recognised as ancestral to all more advanced vascular plants. The last major evolutionary advance among the vascular plants was the emergence of flowering plants (the angiosperms) about 140 million years ago. But the fossils left no clue as to their ancestors. The fossil records also indicate that nearly all the living orders of angiosperms and most of the characters of their modern-day representatives evolved by then.

The continuous change of a character within an evolving lineage is termed as **evolutionary trend**. A lineage is an evolutionary sequence, arranged in linear order from an ancestral group to a descendant group. The number of trends in any lineage is, therefore, same as the number of characters evolving. A trend may be progressive (a general increase in size of organs) or retrogressive (a general degeneration and loss of organs). The evolution of horses (Fig. 3.18) as

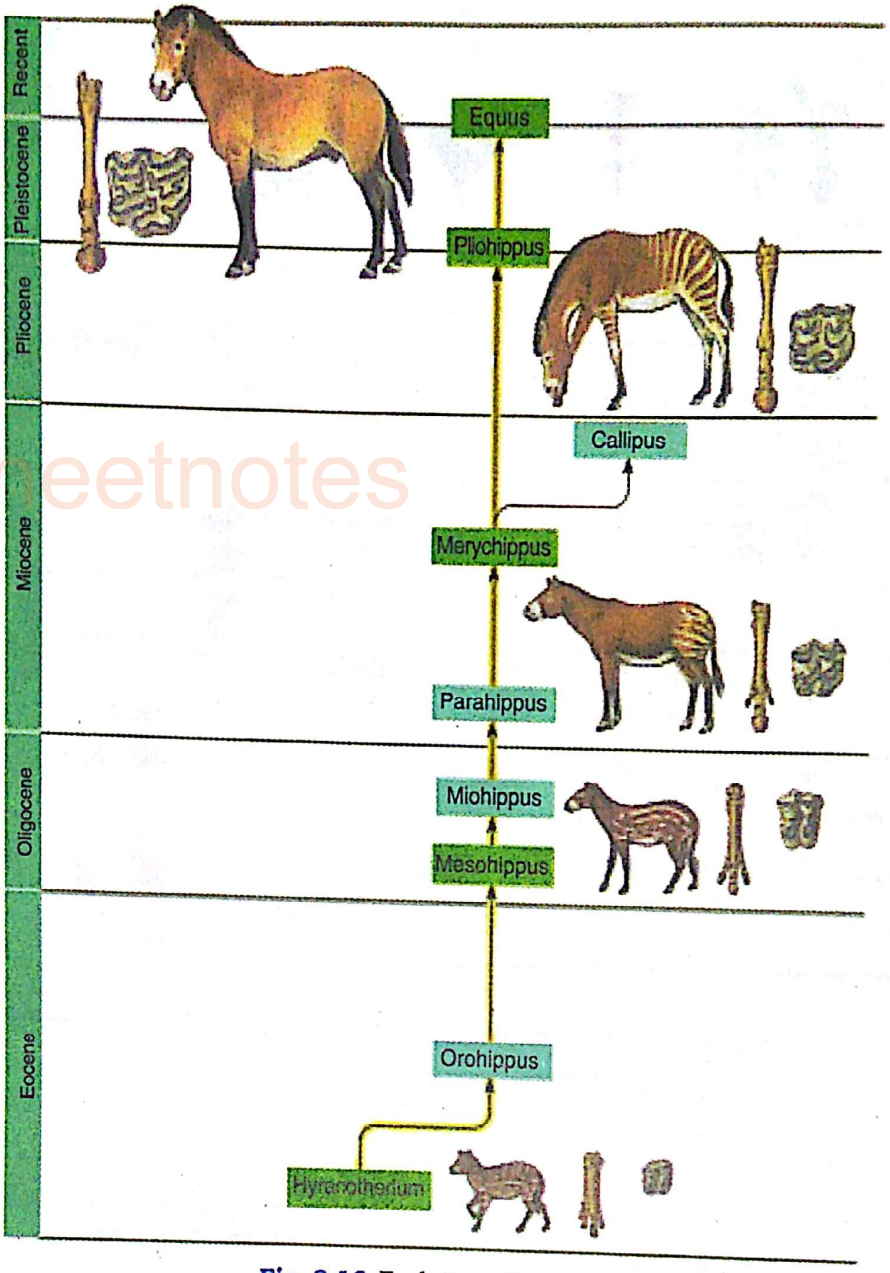

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Fig. 3.18 *Evolution of horses*

evidenced by the fossils reveal a number of important trends. Careful studies of fossils of horses show that the evolutionary trend among horses was not always universal throughout the family. Direction and speed of their evolutionary trend are subject to much variation. The following list identifies the major evolutionary trend of horses:

(i) General increase (with occasional decrease) in size.
(ii) Progressive loss of toes.
(iii) Lengthening of toes that are retained.
(iv) Lengthening of limbs in general.
(v) Enlargement of brain (especially cerebral hemisphere).
(vi) Increase in height.
(vii) Increase in the complexity of molar teeth and an enlargement of the last two and, eventually, the last three premolars until they came to resemble molars.

3.5 THEORIES OF EVOLUTION

An overwhelming suggestion that life had evolved along with evolution of Earth gained currency toward the end of eighteenth century. But the only really pre-Darwinian advocate of evolution was Jean Baptiste Lamarck (1744-1829), a French biologist.

Lamarck's Theory of Evolution or Lamarckism

Lamarck published his theory of evolution in 1809, the year Charles Darwin was born. By comparing the contemporary species of his time to fossil records, Lamarck could arrange the fossils in the chronological series of order from older to younger forms. He could also recognise several lines of descent or lineages of the modern species. Based on his observation, Lamarck proposed that variations among organisms originate because of response to the needs of the environment. Moreover, this ability to respond in a particular direction guides a trait's adaptation. Thus, Lamarck placed fossils in the evolutionary context and stressed on adaptation as means for evolutionary modification. His theory is often called the ***Theory of Inheritance of Acquired Characters*** or the ***Theory of Use and Disuse of Organ***.

Lamarck's propositions

The following are the four propositions :

(i) Living organisms or their component parts tend continually to increase in size.

(ii) Production of a new organ results from a new need and from the new movement, which this need starts and maintains.

(iii) If an organ is used constantly, it will tend to become highly developed, whereas disuse results in degeneration.

(iv) Modifications produced by the above principles during the lifetime of an individual will be inherited by the offspring, with the result that changes are cumulative over a period of time.

Criticism of Lamarckism

Although many lines of descent, for example, evolution of horse, elephant and other animals, portray the first proposition of Lamarck well, it is not universally true. In many groups the size is reduced during evolution. For instance, among angiosperms, the trees seem to have been primitive and the shrubs and herbs including grasses have evolved from these trees.

The second proposition is false. Can we sprout wings wishing to fly like birds? Actually, the importance of inner want or volition of organisms is not testable by scientific methods.

The third proposition appears to have some truth. Lamarck tried to explain the origin of long neck and high shoulders of giraffe on the basis of this principle. According to Lamarck, as the giraffes continually strained to stretch their necks for browsing leaves of higher levels of plants, their necks grew longer and shoulders grew higher in response to their needs. Other prominent examples in this line of argument include the well-developed biceps muscles of blacksmith as an effect of much effort, well-developed muscles in the pinna of rabbits for receiving sound waves from different directions. Limbless condition of snakes, loss of eyes in the cave animals and flightlessness of Kiwi are a few examples of the effectiveness of 'disuse' as a means of evolution. Still, this proposition has been met with many objections. The eyes of a voracious reader do not increase in size and power with increasing age; the constantly beating heart maintains a constant size through generations.

The fourth proposition seems to be a necessary part if environmentally produced modifications are to have any evolutionary significance.

Much earlier, Weissman (1904) observed that mutilation of tails of mice for as many as 22 generations did not result in the birth of a tailless mouse. Mendel's **Laws of Inheritance** and Weissman's ***Theory of Germplasm*** (1892) resulted in the erosion of Lamarck's concept of inheritance of acquired characters. But in Lamarck's times that concept of inheritance was generally accepted and, indeed, Darwin could not offer any acceptable alternative. Lamarck's theory of evolution is now considered as an erroneous assumption since the acquired characters are not inherited.

In retrospect, Lamarck deserves much credit for his vision of evolution as an explanation of the fossil records and the current diversity of life, emphasis on the great age of Earth and stress on adaptation to the environment as a primary product of evolution.

Darwin's Theory of Evolution

Charles Robert Darwin clearly and convincingly set forth the concept of **natural selection** as the mechanism of evolution. Darwin gave the biological world a master key that unlocked the previous intricacies about evolution.

Darwin returned back to England in October 1836 from his expedition with the conviction that all organisms, including humans, are modified descendants of previously existing forms of life: the idea of descent. In 1838, he came across with *An Essay on Population* by Thomas Rev. Malthus (1766-1834) which was published in 1799.

Malthus expressed that famine, war and disease inevitably control the population growth. Darwin noticed the point underscoring conflict between limited resources of population and its continued reproductive pressure. It thus occurred to Darwin that, like in humans, competition exists among all living things.

Darwin assembled data relevant to the mechanism of evolution. He became fascinated by the extensive conscious human efforts in domesticating wild plants and animals since the remote past. Throughout ages humans have been a powerful agent in modifying wild species to suit their own requirements. By conscious selection, the breeders have successfully produced cows that give increased amount of milk. Also we have perfected the toy-like Shetland pony, the Great Dane dog, the sleek Arabian race horse and vast number of cultivated crops and ornamental plants. Also, many crop plants like broccoli, cabbage, cauliflower, etc. have been produced through selective breeding (Fig. 3.19).

The various breeds of fowl ranging from the ceremonial cocks (the Japanese Onaga-dori) to the broiler, leghorns are all derived from a single jungle fowl, *Gallus gallus*. From his inquiries on breeding domesticated plants and animals, Darwin obtained clear evidence for selection, in this case artificial selection. The breeders selected and perpetuated those variant types that interested them or seemed to be useful to them. Darwin himself raised many types of pigeons (Fig. 3.20) by artificial selection.

The analogous process in nature, Darwin called natural selection, which he believed to proceed too slowly to observe. He argued that if so much change can be achieved by artificial selection in a short period of time, then natural selection should be capable of considerable modifications of species over hundreds and thousands of generations. He postulated that natural selection operating in varying conditions over a vast span of time could account for entire diversity of life.

Although Darwin consolidated all his ideas of natural selection he failed to gather enough courage to publish his 'Essays'. Interestingly, in June 1858, Darwin received a brief essay from Alfred Wallace (1823-1913), a naturalist from Dutch East Indies, who was working on Malay Archipelago (present Indonesia). The essay was entitled *On the Tendencies of Varieties to Depart Indefinitely from the Original Type*. Surprisingly, Wallace, who independently conceived the idea of natural selection in a flash of insight during a sudden malarial fever, was also inspired by Malthus's essay on population. To prevent Darwin losing his priority in applying the

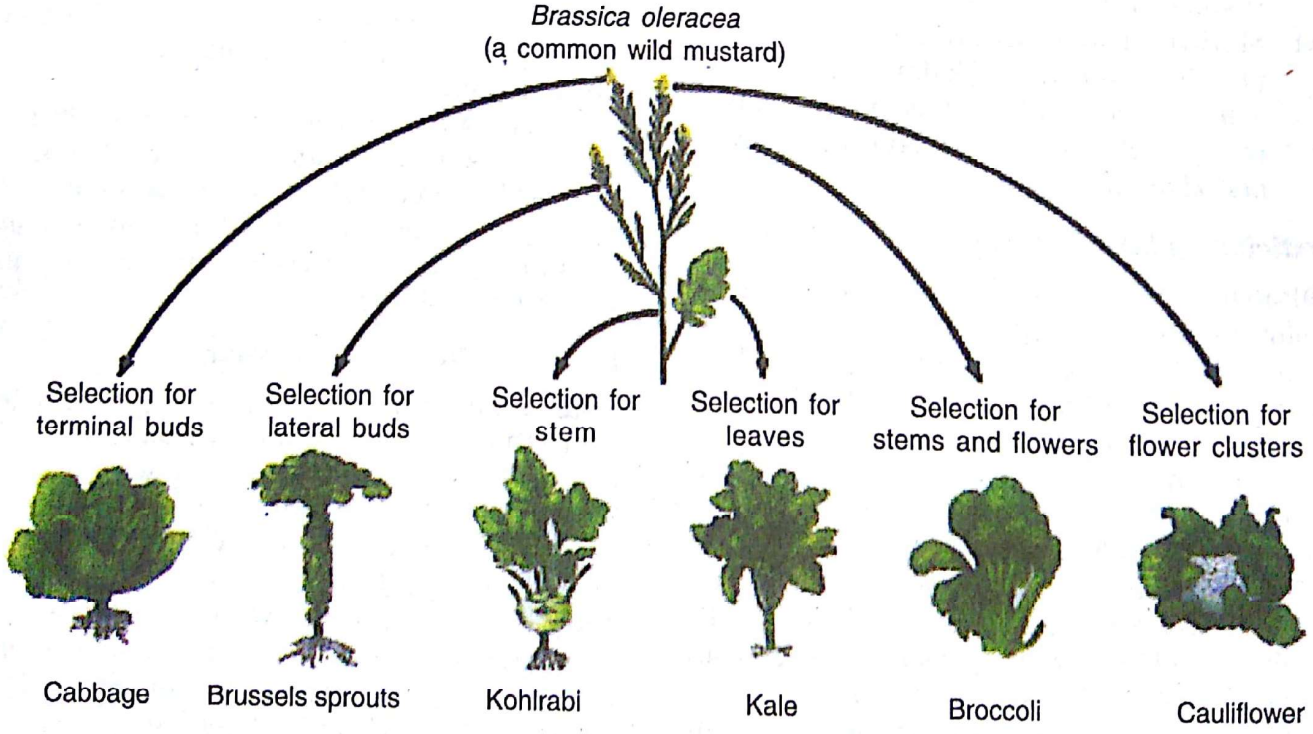

Fig. 3.19 *Some crop plants produced by selective breeding*

Fig. 3.20 *Some of Darwin's pigeons produced by artificial selection*

mechanism of natural selection to evolution, his friends Charles Lyell (1797-1875) and Joseph Dalton Hooker (1817-1911) arranged for short papers on the topic by both authors. On 1 July 1858, Wallace's essay and a portion of Darwin's manuscript were read simultaneously before the Linnaean Society of London.

Darwin then laboured for eight months to abridge his voluminous notes into a single book, which was published in November 1859. The full title of his monumental treatise was *On the Origin of Species by Means of Natural selection: The Preservation of Races in the Struggle for Life.* Darwin actually devoted little space to the origin of species. Rather he concentrated on how populations become well adapted to their local environments through natural selection.

The Principle of Natural Selection

As a whole the principle of natural selection stems from five important observations and three inferences (Ernst Mayr, 1982) that logically follow from them (Table 3.2). Natural selection is this differential success in reproduction and its product is adaptation of organisms to their environment.

Even if the advantages of some variations over others are slight, the favourable variations will accumulate in the population after many generations of being disproportionately perpetuated by natural selection. Thus natural selection occurs through an interaction between the environment and the variability inherent in the population.

Weakness of Darwinism

Although Darwin recognised that the process of evolution is inseparably linked to the mechanism of inheritance he failed to explain the basis of variation and the mode of transmission of the variants to the next generation. In 1868, Darwin put forward his own theory of inheritance, the ***Theory of Pangenesis***. According to this theory, every organ of the body produce minute hereditary particles, called pangenes or gemmules for example liver gemmules from liver, leg gemmules from leg, and so forth. He thought the gemmules were carried through the blood from every organ of the body and were collected together into the gametes. However, August Weismann's ***Theory of Germplasm*** (1892) established that the germ (sex) cells are set apart from other body (somatic) cells early in the embryonic development. Therefore, only changes in the germplasm, can affect the characteristics of future generations. In fact this theory of germplasm refuted Darwin's theory of pangenesis.

de Vries' Theory

Mendel's laws of inheritance paradoxically placed Darwinism in a temporary eclipse. He recognised inheritance as particulate. Nevertheless, he left the question regarding the source of origin of variation unanswered. It was Hugo de Vries (1848 - 1935) who first made a fruitful attempt to answer the questions, not clarified by Mendel. In 1901, he proposed the ***Mutation Theory*** on the basis of his observation on the wild variety of evening primrose (*Oenothera lamarckiana*).

He applied the term spontaneous mutation to discontinuous (discrete) variations that arise all on a sudden. Animal breeders and also Darwin noticed the occurrence of discrete mutations but they called them "sports" and did not consider them of much importance. de Vries expanded these observations into a complete theory of evolution by mutation. For de Vries, it was these

Table 3.2 Natural Selection Stems from Five Important Observations and Three Inferences

Observation	Inferences
1. All species have such great potential of fertility that their population size would increase exponentially if all individuals that were born reproduals succesfully	
2. Most populations are normally stable in size, except for seasonal fluctuations.	
3. Natural resources are limited.	(a) Production of more individuals than the environment can support leads to a struggle for existence among individuals of a population, with only a fraction of offspring surviving each generation.
4. Individuals of a population vary extensively in their characteristics; no two individuals are exactly alike.	
5. Much of this variation is heritable.	(b) Survival in the struggle for existence is not random, but depends in part on the hereditary constitution of the surviving individuals. Those individuals whose inherited characteristics fit them best in their environment are likely to leave more offspring than less fit individuals. (c) The unequal ability of individuals to survive and reproduce will lead to a gradual change in a population with favourable characteristics accumulating over the generations.

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mutations that control evolutionary changes and are more important than natural selection. He dismissed continuous variation among individuals, as proposed by Darwin, as inconsequential and largely nongenetic.

Salient Features of de Vries' Theory

The following constitute the major features of de Vries's theory:

(i) New elementary species originate as a result of large discontinuous variation that appear suddenly and attain full constancy at once.

(ii) The same variations may occur in a large number of individuals.

(iii) Mutations are recurring so that the same mutants appear again and again, thereby increasing the chance of selection by Nature.

(iv) Mutations take place in all directions because it means either gain or loss of any character.

(v) Mutability is something fundamentally different from the fluctuating variability of Darwin, which is small and directional.

Criticism of de Vries' Theory of Mutation

de Vries formulated an extremely important theory that now stands well established. Yet the material on which he worked and on which his theory was based was abnormal. In fact, *Oenothera lamarckiana* is of hybrid nature and the plant itself contains anomalous type of chromosome behaviour. Mutations as conceived by de Vries were mainly chromosomal aberrations that affect the number of chromosomes and are unstable. Our present knowledge of Darwinism reveals that mutations are incapable of introducing new genes and alleles into a gene pool (the sum total of the genotypes or alleles of individuals of a population). This is because natural selection remains inoperative unless there is any change in the gene pool.

Whereas Lamarck, Darwin and others had emphasised the adaptive nature of variation, de Vries stressed on its randomness. While the former scientists highlighted the importance of small and gradual differences in evolution, de Vries focussed on abrupt and often drastic changes as cause of origin of species. Biologists who supported Darwin contended that evolution resulted from gradual fluctuating inheritable variations over a long series of generations. de Vries stated that new species could arise by sudden jump, often called saltation or through a single step large mutation (macrogenesis). However, the debate between gradual evolution and evolution by saltation still remains unresolved.

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3.6 MODERN VIEWS OF DARWINISM

Although de Vries postulation initially reinforced suspicion of the contemporary geneticists for natural selection, ultimately it paved the way toward proper understanding of Darwinian view of evolution. Underlying the process of change and diversification of organisms through time (evolution on the grand scale of geological time, **macroevolution**) there are changes in the hereditary materials (evolution at the genetic level, **microevolution**). During the first part of twentieth century incorporation of population genetics into the studies of evolution led to the emergence of a comprehensive theory of evolution, called modern synthesis. The modern synthesis emphasises the importance of populations as the units of evolution and the central role of natural selection as the most important mechanism of evolution.

Population Genetics and Evolution

Population genetics is the study of the frequencies of genes in populations. It represents an application of Mendelian genetics to Darwinian natural selection. A population may be defined as all individuals of the same species occurring in the same area at a particular time. Evolution occurs within populations as the relative frequencies of different variations of DNA change over time. If there are two forms of a specific enzyme in a population and if the relative frequencies of the individuals possessing each form of enzyme changes then certainly there is evolution.

The well known **Hardy-Weinberg Equilibrium** (theorem) or **Principle**, which was proposed by G. H. Hardy and W. Weinberg, independently, in 1908, defined the genetic structure of a non-evolving population. They observed that hereditary conservation of genes is the characteristic of a population. According to Hardy-Weinberg law, under certain conditions of stability allelic frequencies remain constant from generation to generation in sexually reproducing organisms. This means that, if all other factors remain constant, the frequency of particular genes and alleles will remain constant in a population through generations. This kind of stability at the genetic level is called genetic equilibrium.

Hardy-Weinberg Principle gives the geneticists a tool to determine when evolution is occurring. Population geneticists use this principle to calculate a starting point allele frequency and then compare it to frequencies measured at some future time. The amount of deviation between observed frequencies and those predicted by Hardy-Weinberg Principle indicates the degree of evolutionary change. Therefore, evolution occurs when the equilibrium is upset. In other words, evolution is a departure from Hardy-Weinberg Equilibrium.

Sources of Variation

The individuals of a population share some important features but differ from one another in numerous ways. In fact, with rare exceptions, no two members of a population are exactly alike. For a population to evolve, its members must possess variation, which is the raw material on which agents of evolution act. Variation can be observed both at the phenotypic level and at the genetic level. Phenotype is the physical expression of genes of an organism. The genetic constitution that governs a trait (heritable characteristic) of an organism is called genotype. Natural selection can act on genetic variation only when it is expressed in the phenotype. Hence, evolution requires genetic variation. In the example of peppered moth, *Biston betularia*, if there were no dark moth, the population of moths could not have evolved from most light to most dark forms. In order to continue evolution, there must be mechanisms to create increase or decrease in genetic variation.

Evolutionary agents are the forces that change the allele and genotype frequencies in a population. In other words, they cause deviations

from Hardy-Weinberg Equilibrium. Five basic processes affect the Hardy-Weinberg equilibrium and cause variation at the genetic level. These are **mutation, recombination, gene migration, genetic drift** and **natural selection.**

Mutation

To Hugo de Vries, a mutation was sudden heritable change. These mutations are random (indiscriminate) with respect to the adaptive needs of organisms. Most mutations are harmful or with no effect (neutral) on their bearers. If the environment changes, however, previously harmful or neutral alleles may become advantageous. Mutation rates are very slow for most loci that have been studied. Usually, one mutation in a million occurs. Nonetheless, these mutation rates are sufficient to create considerable genetic variation.

By an ingenious experiment Joshua Lederberg and Esther Lederberg (1952) were able to show that there are mutations which are actually preadaptive. These kinds of mutations are regarded as advantageous mutations. They appear without exposure to the environment in which they would be advantageous to the organism. Actually, the preadapted mutations express themselves only after exposure to the new environment to which the organisms are to adapt themselves. The new environment does not induce their formation; it only selects the preadaptive mutations that occurred earlier.

They used the techniques of replica plating (Fig. 3.21). From a culture of *Escherichia coli* derived from a single cell, they spread cells onto a 'master'agar plate. Each cell gave rise to a distinct colony. They then placed a velvet cloth on the master plate and touched it to a new plate with medium containing the antibiotic penicillin. By this method, they transferred some cells from each colony to the replica plate, in the same spatial relationship as the colonies from which they had been taken. A few colonies appeared on the replica plate, having grown from penicillin resistant mutant cells. When all the colonies on the master plate were tested for penicillin resistance, only those colonies that had been the source of penicillin resistant cells on the replica plate displayed resistance. This proved that mutations had occurred before the bacteria were exposed to penicillin.

Mutation can restore to populations' alleles that other evolutionary agents remove. Thus,

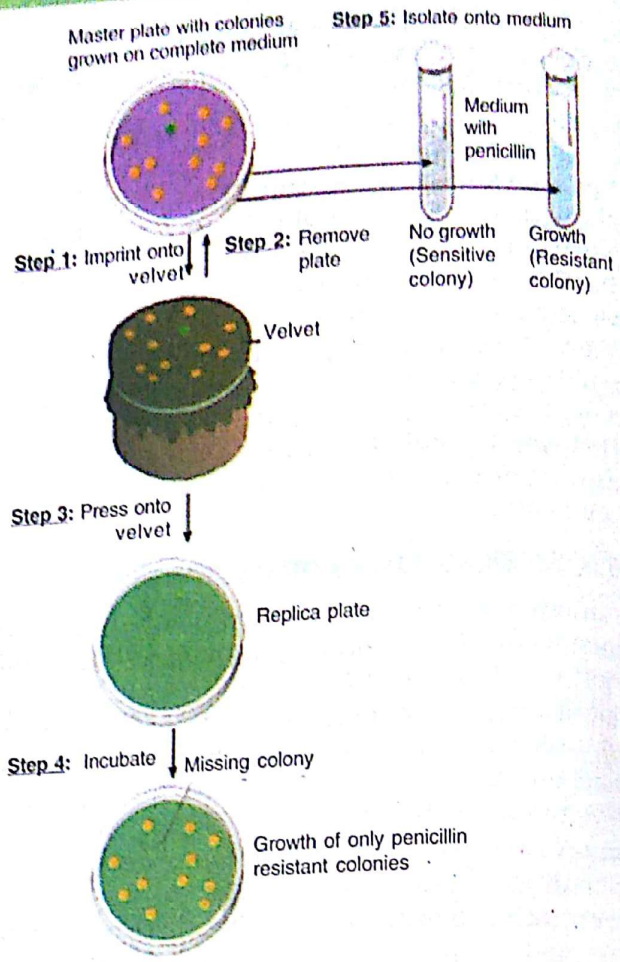

Fig. 3.21 Replica plate experiment of Lederberg and Lederberg

mutations both create and help maintain variations within population. Mutations also introduce new genes and alleles into the gene pool. Gene pool is the sum total of all the genotypes or alleles (Fig. 3.22) found in a population.

It is the gene pool that evolves as new genes or variants of genes, i.e. alleles, are added or removed. This variability of genes or alleles in a gene pool, then, is the raw material for evolutionary change. Over hundreds of generations, the accumulation of many mutations may add up to large-scale changes.

Recombination

During meiosis, crossing over causes reshuffling of gene combinations which provides new combinations of existing genes and alleles. This is the essence of recombination. It may bring together the alleles that arose at different times and places. Recombination can occur not only

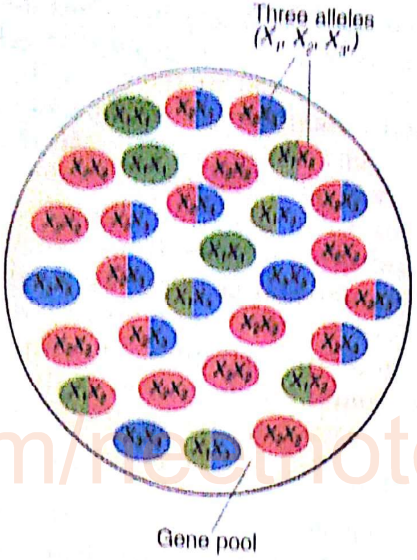

Fig. 3.22 *Alleles in a Gene pool*

between genes but also within genes resulting in the formation of a new allele. As it adds new alleles and combinations of alleles to the gene pool it is an agent of evolution.

Gene Migration

Because few populations are completely isolated from other populations of the same species, usually some migration between populations takes place. Therefore, members of a population may enter a new population due to migration. If the migrating individuals breed within the new population, the immigrants will add new alleles to the local gene pool of the host population. This is called gene migration. If the immigrated species is closely related with the host species, fertile hybrids may result from interspecific mating. These hybrids, then, can carry genes from species to species. The addition or removal of alleles when individuals enter or exit a population from another locality results in gene flow. Therefore, migration - emigration or immigration results in the diffusion of genes into populations. Unrestricted gene flow decreases the differences between the gene pools and, consequently, reduces the distinction between separated populations.

Genetic Drift

The random changes in the allele frequency occurring by chance alone are called genetic drift. Drift is a binomial sampling error of the gene pool. This means that the alleles that form the gene pool of the next generation are a sample of the alleles from current generation. Sampling errors (chance) often lead to the elimination of certain alleles and fixation of others, thus, reducing the genetic variability of the population. In a small population, a chance event (e.g. snow storm) may increase the frequency of a character that has little adaptive value. Genetic drift can cause dramatic changes in the allele frequencies in a population derived from small bands of colonisers, called *founders*, to a new habitat. Often their phenotypes quickly become different from the parental population, sometimes forming a new species. Such an effect is called the **founder effect**. Also population crashes leading to drastic reduction in population size can cause change in allele frequencies. Given the fact that the existing gene pool is limited, population crashes retard the ability of the population to reestablish its former richness. Such reduction in allele frequencies is called a genetic bottleneck (Fig. 3.23), which often prevents the species from reversing its path of extinction.

Natural Selection

Natural selection is the most critical evolutionary process that leads to changes in allele frequencies and favours or promotes adaptation as a product of evolution. Also it keeps the disharmonising effects of the other processes, which are not oriented to adaptation, in check. Well-adapted individuals (pre-existing classes of genetic variants in the gene pool) generally survive longer and reproduce more offspring. Through such differential reproduction adaptive alleles are selected for transferring to next generation and so increase in frequency over generations. Alleles that are less adaptive, in reproductively less successful individuals are not selected and so decrease in frequency through generations. In Darwinian terms, survival and fertility mechanisms that affect the reproductive success or promote differential reproduction are known as selection. In modern terms, selection is the consistent differences in the contribution of various genotypes to the next generation. To bridge the two views we may say that reproduction leads to differential contribution of genotypes to the gene pool of the next generation.

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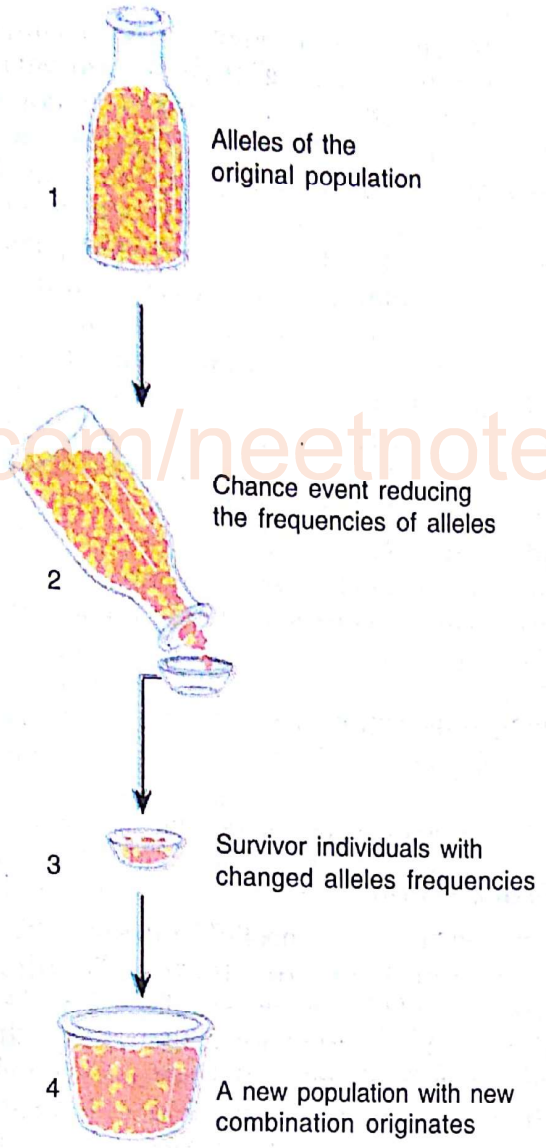

Fig. 3.23 *Explaining bottleneck effect -frequencies of two alleles are represented by red and yellow colours.*

Natural selection causes allele frequencies of a population to change. Depending upon which traits are favoured in a population, natural selection can produce any one of several quite different results (Fig. 3.24). If both the smallest and largest individuals contribute relatively fewer offspring to the next generation than those closer to the average size do, stabilising selection is operating. **Stabilising selection** reduces variation but does not change the mean value. Rates of evolution are typically very slow because natural selection is usually stabilising.

If individuals at one extreme of the size distribution (e.g. the larger ones) contribute more offspring to the next generation than the other individuals do, then the mean size of individuals in the population will increase. In this case, **directional selection** is operating. If directional selection operates for many generations, an evolutionary trend within the population results.

When natural selection simultaneously favours individuals at both extremes of the distribution, **disruptive selection** is operating. This kind of selection is rare. When it operates, individuals at the extremes contribute more offspring compared to those in the centre, producing two peaks in the distribution of a trait.

3.7 GENETIC BASIS OF ADAPTATION

Darwin visualised that evolution takes place by natural selection. Also we have learnt that an evolutionary change bears a genetic basis for causing hereditary variation. It is this genetic variation that helps, if favoured by natural selection, the organisms to adapt to a particular environment. Thus, any evolutionary adaptation must have its basis at the genetic level. Any adaptation without genetic basis has no evolutionary significance.

Industrial Melanism

A classic example of natural selection in the wild is the case of peppered moth, *Biston betularia*, which lives in all parts of England. This moth shows cryptic coloration (Fig. 3.25) with two phenotypes, light (gray) and dark (black). During 1850 to 1950, biologists found the black variety became more and more common and the light form scarcer, particularly in the industrial cities (Birmingham, for example). This change in the population of peppered moth is, in itself, evolution. Biologists proposed that before industrial pollution, the typical light form of peppered moth succeeded to camouflage well against the pale tree trunks on which they used to rest during daytime. Due to industrial smoke and soot the pale tree trunks became more and more blackened. As a result the light moths stood out in contrast to its background, increasing the possibility of being easily detected and eaten by their predators, such as birds, in much greater number than the dark melanic variety. Decrease in the number of light moths and increase in the number of dark variety was the ultimate result. Therefore, evolution favoured

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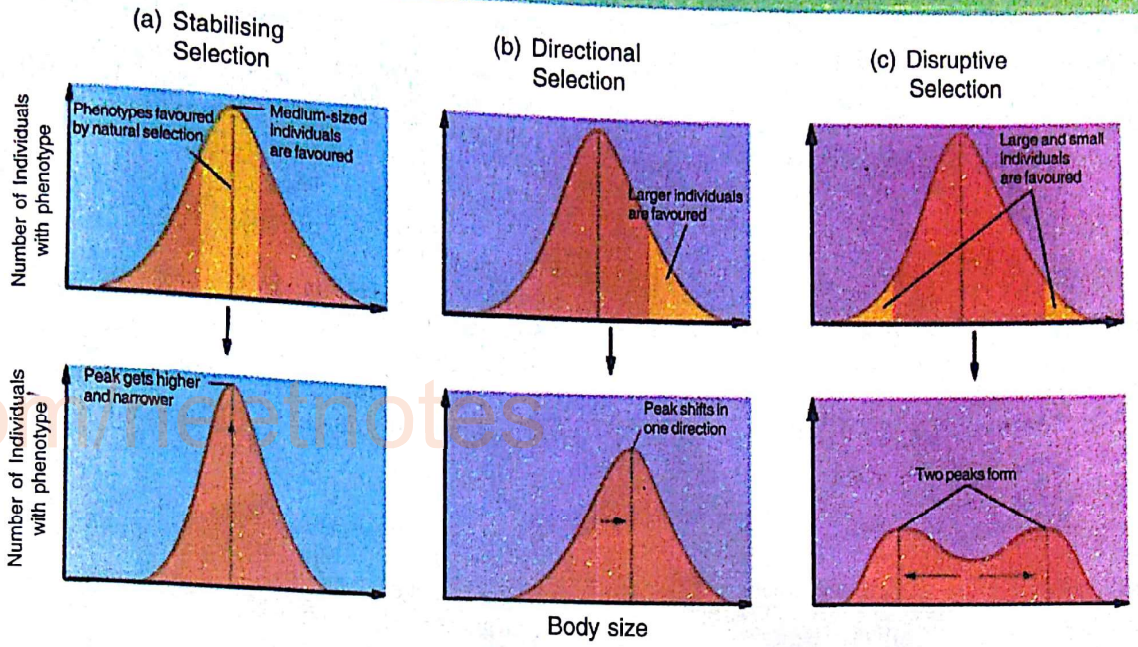

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Fig. 3.24 *Effect of natural selection on a variable trait (body size) - (a) Stabilising selection (left upper and lower set), (b) Directional selection (middle) and (c) Disruptive selection (right)*

the melanic moths to reproduce more successfully for their adaptation in the polluted areas of England. Evolution of darker form in response to industrial pollution is known as industrial melanism.

Barnard Kettlewell, a British ecologist, tested the hypothesis in the 1950s by rearing populations of peppered moths with equal numbers of dark and light individuals. He released these moths in two sets - one in the woods of Birmingham (polluted area), the other in Dorset (unpolluted area). In the polluted area Kettlewell could recapture 19 per cent of light moths and 40 per cent of dark ones. In the unpolluted area he could recapture only 12.5 per cent of the light moths and 6 per cent of dark moths. This result focuses the patterns of differential survival of *Biston betularia* in the polluted and unpolluted areas. The data of Kettlewell and others suggested that sooty areas offer great protection to melanic forms. The increased proportion of dark peppered moth in the relatively more polluted (industrial) area has its basis in the increased frequency of a dominant gene in industrial area. Whereas in the unpolluted area the frequency of the gene responsible for light-coloured moths have more selective advantage. Interestingly, passage of clean air legislation in Britain in 1956 has reduced industrial smoke and sulphur dioxide in many formerly polluted areas. Consequently, the frequency of melanic forms of peppered moth and other insects has declined dramatically. Thus, reduction in pollution is now correlated with *reverse evolution*. Moreover, industrial pollution did not eliminate the gene responsible for light-coloured moths completely. Dozens of other species of moths have changed in the same way in industrialised areas throughout Eurasia and North America. Industrialisation has boomed the selection and evolution of dark forms from the mid-nineteenth century.

Natural Selection and Polymorphism

A population sufficiently widespread to occupy many environments may maintain a variety of genotypes, each of which is superior in a particular habitat. A population is called polymorphic for a character if two or more distinct morphs (forms) are represented in high enough frequencies. Polymorphism is extensively represented in humans. One example is ABO blood group. There are four morphs: type A, type B, type AB and type O determined by different genotypes. The persistence of different genotypes through heterozygote superiority is another example of polymorphism. It is meant for preservation of genetic variability through

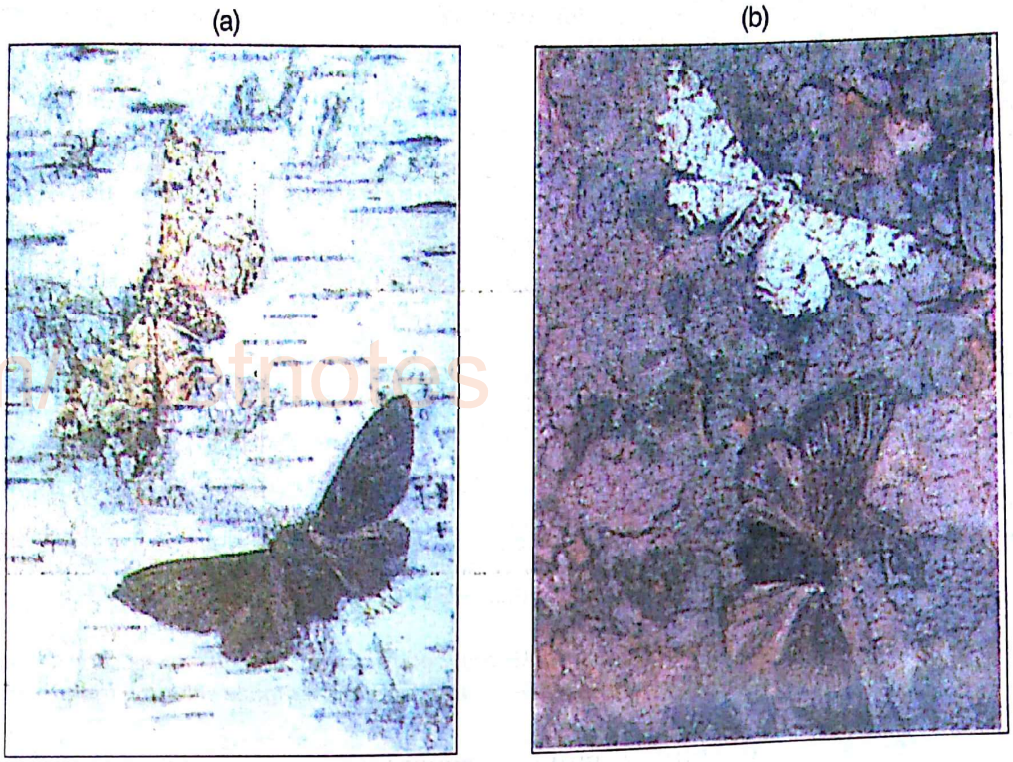

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Fig. 3.25 *Cryptic coloration of Biston betularia (a) Light and dark moth on a tree trunk in unpolluted area (b) a soot-covered tree trunk in a polluted area*

heterozygote selection. This is called **balanced polymorphism**.

Usually, a gene locus is considered polymorphic if at least two alleles are present, with a frequency of at least one per cent for the second most frequent allele. One prominent example of polymorphism that overdominance causes is sickle cell gene in human populations. Variation at a single locus determines whether red blood cells are shaped normally or like a sickle. Homozygotes with two normal alleles (Hb^A/Hb^A) or with two alleles for sickle cell (Hb^S/Hb^S) suffer from malarial attack very easily. Besides, the heterozygotes develop anaemia – the very shape of the sickle cells prevents them from carrying normal level of oxygen. But the heterozygotes (Hb^A/Hb^S), who have one copy of sickle cell allele, coupled with one normal allele, enjoy some resistance to malaria; they survive the malarial parasite more successfully than either normal or sickle cell homozygotes. The high incidence of this deleterious gene in human population is due to favourable selection by nature. Moreover, the loss of deleterious recessive genes through deaths of homozygotes (Hb^S/Hb^S) is being balanced by the gain resulting from successful reproduction by heterzygotes. For this reason such selection is called **balancing selection**. It should be pointed out that balancing selection is rare and thus should not be considered as a general explanation for the levels of genetic variations found in natural populations.

3.8 SPECIATION AND ISOLATION

Speciation is the formation of one or more new species from an existing species. The crucial episode in the origin of species occurs when the gene pool of a population is severed from other populations of the parent species and gene flow no longer occurs. Speciation can take place in two modes based on the geographical relationship of a new species to its ancestral species. When a population, formerly continuous in range, splits into two or more geographically isolated populations and form new species, the mode of speciation is called **allopatric speciation**. This can happen by subdivision of

the original population, when a geographical barrier, such as a creeping glacier, a land bridge (e.g. Isthmus of Panama) or ocean or mountain, cuts across a species range. Alternatively, when a small number of individuals colonise a new habitat that is geographically separated from the original range. Recall the examples of Darwin's finches that formed separate species in the Galápagos islands and the Australian marsupials that radiated to form new species. In the second speciation mode, a subpopulation becomes reproductively isolated in the midst of its parent population; this is **sympatric speciation**. So, sympatric speciation is the formation of species within a single population without geographical isolation. The usually quoted example of sympatric speciation comes from **polyploidy**, which is the multiplication of the normal chromosome number. This can happen when chromosomes fail to segregate at meiosis or replicate without undergoing mitosis.

Reproductive isolation may be defined as the existence of intrinsic barrier to the interbreeding of natural populations. Each of these intrinsic barriers is called a reproductive isolating mechanism. According to Mayr (1942), reproductive isolating mechanisms are the *biological properties of individuals which prevent the interbreeding of naturally sympatric populations.*

Reproductive isolating mechanisms may be classified as either premating or postmating. Premating isolating mechanisms act prior to mating. These may be ecological, behavioural and mechanical. Postmating isolation mechanisms act subsequent to mating, preventing the interbreeding populations by removing the hybrids from the gene pool. These mechanisms may be gametic mortality, zygotic mortality, embryonic or larval mortality, hybrid inviability, hybrid sterility and F_2 breakdown. Natural selection tends to favour the organisms that exhibit premating isolation over those that have only postzygotic isolation. Reproductive isolation has its bearing on genetic isolation which does not permit populations to exchange genes. Therefore, the gene pools of different species are isolated from one another. On the contrary, members of the same species share a common gene pool. On this basis, a species can be defined as one or more populations sharing a common gene pool.

Reproductive isolation in the form of hybrid sterility is known since long. In the laboratory or in zoos, hybrids can be produced between species that do not interbreed in nature. Horses and donkeys are two different species; a hybrid, mule, is produced from the mating of a male donkey and a mare (female horse). Similarly, mating between stallion (male horse) and female donkey results in a hybrid called hinny. Both mule and hinny are sterile. There are examples of species, which can produce fertile hybrids in captivity. You might have heard about the famous 'tigons', a hybrid of African lioness (*Panthera leo*) and Asian tigers (*Panthera tigris*), which is fertile. No barrier to hybridisation between these species has evolved during their long isolation from each other. Natural selection has not favoured a reduction in hybridisation for the simple reason that no hybridisation has been possible. Other examples of species that breed in captivity and produce fertile hybrids are mallard (a duck) and the pintail duck, the polar bear and the Alaskan brown bear and the platy and swordtail fishes. But these species do not interbreed at all in natural condition.

3.9 SPECIES CONCEPT

The main goal of **evolutionary taxonomists** is to recognise the basic unit of classification, the species. Taxonomists also try to order the species into as realistic a phylogenetic scheme as possible. They have used various methods to define species. Davis and Heywood define **species** as *assemblage of individuals with morphological features in common and separable from other such assemblages by correlated morphological discontinuities in a number of features.* **Lumpers** tend to combine populations into single species or groups. **Splitters** tend to separate the same populations into different species or groups.

Many biologists have not been satisfied to let species definitions rest primarily on a subjective or qualitative morphological approach. They have, instead, adopted a **biological species concept**. Traditionally, the biological species concept defines a species (*L*: form) as a sexually interbreeding or potentially interbreeding group of individuals separated from other species by the absence of genetic exchange, that is, by reproductive isolation. Members of a sexually

reproducing species are capable of breeding with one another in nature to produce viable, fertile offspring but unable to breed with members of other species. When the two species are morphologically almost identical but do not normally interbreed, such species are called **sibling species**. For example, *Drosophila pseudoobscura* and *D. persimilis*, two species of fruit fly do not cross-fertilise. Biological tests to cross-fertilise morphologically and geographically separated species have led the scientists to unify different groups into single species, called **polytypic species**. For instance, the various species of North American sparrows has been united with the multiple geographical races or sub-species of song sparrow, *Passarella melodia*. However, this concept has some problems. Although most organisms are sexual, there are many organisms that are asexual. For example, all prokaryotes, some protists, some fungi, some plants (such as commercial banana) and some animals. Asexual organism may be assigned to species only on grouping of clones that have the same morphology and biochemistry. The enormous number of species, their varied geographical dispersal and because of the fact that most species are extinct many species can not be differentiated by this criterion of reproductive isolation.

Many authors have proposed an evolutionary species concept in an attempt to circumvent some of the problems of biological species concept. In this concept, species are defined in terms of differences that are not dependent on sexual isolation. The supporters of this concept rather value the 'evolutionary' isolation, of which sexual isolation is only one aspect. According to George Gaylord Simpson (1961), *an **evolutionary species** is a lineage (an ancestor-descendant sequence of population) evolving separately from others and with its own unitary evolutionary role and tendencies*. This concept, for the first time, incorporates change (evolution). Ideally, this method uses morphological, genetic, behavioural, and ecological variables. Yet it does not resolve all the problems intrinsic to species taxonomy. We know that all traits do not evolve at the same rate or in the same sequence. In summary, the difficulties in species taxonomy are inherent in the process of speciation itself.

SUMMARY

The history of life comprises two events — origin and evolution. There is a general consensus that life arose from collections of organic molecules by a series of progressive chemical reactions. Oparin and Haldane suggested that spontaneous generation of early organic molecules might have taken place if the primeval Earth once contained virtually no oxygen and atmosphere was reducing. Geological record shows the presence of such reduced materials as uranite, pyrite, etc. in the sediments, implying that the conditions then were strongly reducing.

Miller obtained significant amount of the simple major organic compounds, such as amino acids and peptide chains in the aqueous sample of the experimental set. Abiogenic synthesis of building blocks of life would thus appear to be an undeniable step in chemical evolution during primeval time on Earth. Aggregates of artificially produced prebiotic molecules, called protobionts, are seen to separate combination of molecules from the surroundings and maintain an internal environment. Most probably, the first genetic code was based on RNA that catalysed its own replication and other chemical reactions. The accumulated products of RNA-catalysed reactions could then participate in other reactions and form structures. DNA probably did not evolve as a hereditary molecule until RNA-based life became enclosed in membranes. Once cells evolved, DNA probably replaced RNA as the genetic code for most organisms.The first living organisms arose in a sea of organic molecules and were, presumably, anaerobes and heterotrophs. However, some of the heterotrophs might have evolved into autotrophs, which produced their own organic molecules by chemosynthesis or photosynthesis. The emergence of photosynthesis was a turning point because this process changed the atmosphere of the Earth and, hence, initiated evolution of life toward its diversities.

The word evolution means to unfold or unroll or to reveal hidden potentialities. Change in the planets and stars in between their birth and death is stellar or cosmic evolution. The matters – elements, and their subatomic particles – change in time. This is inorganic evolution. The changes in the properties of population of organisms or groups of such populations over the course of generations are considered as biological or organic evolution. It is descent with modifications. The branch of biology that involves the study of living systems as they change through time is known as Evolutionary Biology. All theories of evolution, from Darwin to the present day, are permeated with numerous earlier concepts and theories. The ancient Indian texts of philosophy and Ayurveda deal with the origin of life. Manu's texts in Sanskrit, Manu Samhita or Manu-smriti (200 AD) also make a mention about evolution. Several different lines of evidence convinced Darwin and his contemporaries that modern organisms arose by evolution from more ancient forms.

Fossils are the preserved remains, or traces of organisms that lived in the past. The fossils contained in layers of sedimentary rock reveal a history of life on Earth. The rocks are dated by measuring the degree of spontaneous decay of certain radioactive isotopes. As a result, the actual age of rocks and their fossils are now possible to find out and the geological time scale has been prepared. Fossils have been found linking all major groups of vertebrates.

Lamarck's theory of Inheritance of Acquired Characters or Theory of Use and Disuse of Organ held that living organisms tend continually to increase in size and new organ results from a new need and from the new movement. All the experimental results failed to coincide with those of repeat experiments performed by different scientists, Mendel's laws of inheritance and Weissman's theory of germplasm (1892) resulted in the erosion of Lamarck's concept of inheritance of acquired characters. Mutations as conceived by de Vries were mainly chromosomal aberrations that affect the number of chromosomes and are unstable. Incorporation of population genetics into the studies of evolution led to the emergence of modern synthesis, which emphasises the importance of populations as the units of evolution and the central role of natural selection as the most important mechanism of evolution. Evolution occurs within populations as the relative frequencies of different variations of DNA change over time. On the contrary, Hardy and Weinberg observed that hereditary conservation of DNA (genes) is the characteristic of a population. Recombination brings together the alleles that arose at different times and places. Natural selection leads to changes in allele frequencies and favours or promotes adaptation as a product of evolution. Also it keeps the disharmonising effects of the other processes, which are not oriented to adaptation, in check. In modern terms, natural selection is the differential reproduction leading to differential contribution of genotypes to the gene pool of the next generation.

Speciation is the formation of one or more new species from an existing species. It occurs when the gene pool of a population is severed from other populations of the parent species and gene flow no longer occurs. Taxonomists define species as an assemblage of individuals with common morphological features and separable from other such assemblages by correlated morphological discontinuities in a number of features. Biological species concept defines a species as a sexually interbreeding or potentially interbreeding group of individuals separated from other species by reproductive isolation.

EXERCISES

1. Name two major events of the history of life. Briefly state the major theories of origin of life. Which one of them has scientific basis?
2. What is cosmology? Name the major theories of origin of the universe. Briefly narrate the mostly accepted one.
3. State the hypothesis of Oparin and Haldane about the primeval Earth condition. What do you understand by Haldane's hot dilute soup? State its significance.
4. Summarise Miller's simulation experiment for organic synthesis. Comment on its efficacy.
5. What is "protobiont"? Name the various types of protobionts conceived by different scientists. Mention their characteristics and drawbacks.

6. What is meant by the term evolution? Distinguish between cosmic evolution and organic evolution. What is evolutionary biology?
7. What is biogeography? How Darwin succeeded to use the evidence from biogeography in favour of evolution?
8. Explain convergent evolution with a suitable example.
9. Distinguish between homologous and analogous organs. Give examples of homologous organs found in animals and analogous organs found in plants.
10. What is a vestigial organ? Give some examples of vestigial organs. State their significance.
11. Elucidate Baer's law and its significance.
12. What is a fossil? Discuss the significance of fossils in evolution.
13. Enumerate Lamarck's propositions about evolution. Describe the examples given by Lamarck in support of the theory of use and disuse.
14. Discuss the importance of artificial selection in the derivation of the concept of natural selection.
15. Give a tabular representation of the principle of natural selection.
16. Mention the salient features of de Vries's theory of mutation. Comment on the generalistion made by him on evolution.
17. Distinguish between microevolution and macroevolution. Narrate the significance of population genetics in evolution.
18. What is variation? Name the basic processes that cause variation among organisms. Discuss the role of migration in evolution.
19. Define genetic drift. How does it produce founder effect and genetic bottleneck.
20. What is natural selection in modern terms? Elucidate the three different effects of natural selection on variation.
21. Write notes on
    (i) Replica plating
    (ii) Gene flow
    (iii) Industrial melanism
    (iv) Balancing selection
    (v) Hybrid sterility
    (vi) Allopatric and Sympatric speciation
    (vii) Biological species concept
    (viii) Evolutionary species concept

22. Fill in the blanks
    (i) About 4.6 billion years ago the Earth consisted of a solid _____ and a gaseous _____.
    (ii) In the primeval Earth, solar radiation, lightning and _____ served as the energy sources for _____ reactions of organic synthesis.
    (iii) In the primeval Earth, the organic molecules accumulated in _____ because their _____ was extremely slow in the absence of any life or enzyme catalysts.
    (iv) The first living organisms were, presumably, _____ and heterotrophs, some of which might have evolved into _____ autotrophs.
    (v) The earliest evidence of life appears in the microfossils of _____ _____ that appeared 3.3 to 3.5 billion years ago.

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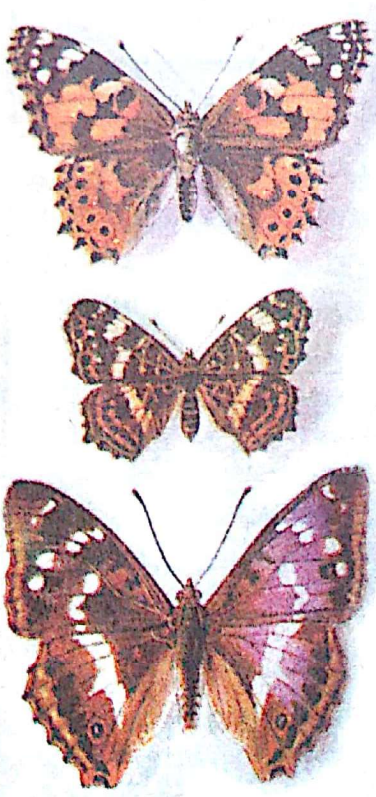
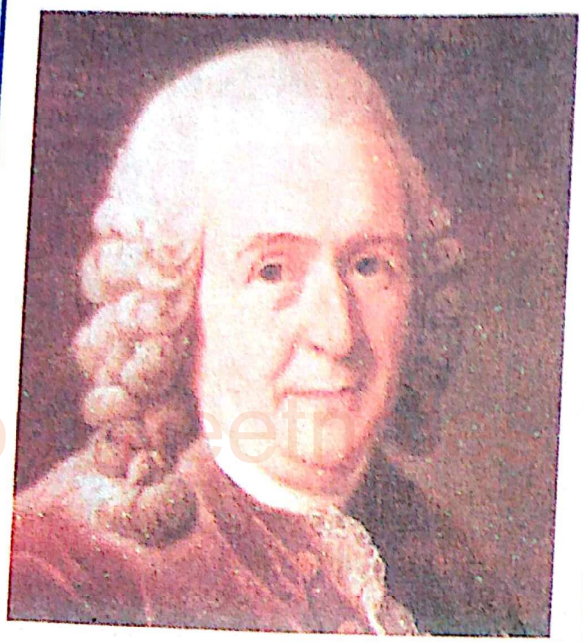
UNIT
TWO



Diversity of Life

In the previous unit you have learnt that all organisms share many fundamental and common characteristics. Also you got a glimpse of the evolution and its role in diversity of life. The living world comprises millions of different kinds of organisms. You must be familiar with different types of plants and animals in your surroundings. To study them is to know the diversity of life forms. Studies on the diversity of life require that the organisms should be given names. You can group some of the organisms into basic units as teaks, oaks, insects, cattle, and so on. The fact remains that most of the organisms have no common name and even if it exists, it is unreliable and often confusing. Recognising and interpreting similarities and differences among organisms is easier if the organisms are classified into groups that are ordered and ranked. About 300 years ago, scientists first began to develop an overall classification system of organisms and to produce comprehensive works about them. Thousands of newly identified species are being added to the list of living organisms each year. It is believed that total number of species that exist on Earth ranges between 9-52 millions. This unit will help you to know about the various aspects of biological classification.



CAROLUS LINNAEUS

(1707-1778)

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Carolus Linnaeus (Karl von Linne) was born in Sweden. As a child, he developed an interest in plants. On persuasion he joined the medical school, though his parents wanted him to be a minister. Medical course being closely related to plants then, Linne continued with the collection, study and description of plants. After completing medical degree in 1739, he went to Holland and became the personal physician of a wealthy government official. There he also studied and described all the plants of his employer's garden. At the age of 22 , he published his first paper on sexuality of plants. Later he published 14 treatise and also brought out famous ***Systema Naturae*** *from which all fundamental taxonomical researches emanated. His system of classification was a simple scheme for arranging plants for identifying them again. Karl von Linne with his lectures and publications in Latin became Carolus Linnaeus.*

Chapter 4

SYSTEMATICS

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There are millions of organisms – plants, animals and microbes. Each one is different from the other in one way or the other. More than one million of species of animals and more than half a million species of plants have been studied, described and provided names for identification. Thousands are still unknown and are yet to be identified and described. It is practically impossible to study each and every individual. Also, it is difficult to remember their names, characters and uses. However, biologists have devised techniques for identification, naming and grouping of various organisms.

4.1 WHY STUDY CLASSIFICATION?

The art of identifying distinctions among organisms and placing them into groups that reflect their most significant features and relationship is called **biological classification**. The purpose of biological classification is to organise the vast number of known plants and animals into categories that could be named, remembered and studied. As a result, the study of one organism of a group gives us the idea about the rest of the members of that particular group. Scientists who study and contribute to the classification of organisms are known as systematists or taxonomists, and their subject is called **systematics** (*Gk. Systema:* order or sequence) or **taxonomy** (*Gk.Taxis:* arrangement; *nomos*; law). Generally, the terms such as classification, systematics and taxonomy are used interchangeably but some taxonomists, like G.Simpson(1961), relate them to separate fields. He considers systematics as the study of diversity of organisms and all their comparative and evolutionary relationships, based on comparative anatomy, comparative ecology, comparative physiology and comparative biochemistry. He regards **classification** as a subtopic of systematics that deals with ordering of organisms into groups and taxonomy as the study of principles and procedures of classification. Be that as it may, they are quite interrelated fields and, henceforth, will be used interchangeably.

Taxonomic knowledge about the organisms is based on their form and structure (morphology), cell (cytology), development process (embryology), remnant of the past organisms (fossils) and ecological relationships. This knowledge gained through taxonomy is assembled for future use not only for the biologists but also for others working in the fields of medicine, agriculture and forestry or industry and so on. In order to understand the living world, especially its diversity, it is essential to make inventory of organisms with correct identification and names. It further facilitates classification of organisms based on their similarities or evolutionary relationships (phylogeny). Moreover, the biologists considerebly depend on studies of fossils for drawing evolutionary relationships among the organisms. You might recall here the impact of taxonomy on different aspects of biology in section 'Scope of Biology' (Chapter 1).

4.2 HISTORY OF CLASSIFICATION

Plants and animals were present on this planet before the humans evolved. However, the humans as the advanced species with communication through language, started using plants and animals for their basic needs. They gave names to the plants and animals, noted their features and even classified them in their own language.

Our Vedic literature (2500 BC to 650 BC) recorded about 740 plants and 250 animals.

Chandyogya Upanishad classifies animals. Post-Vedic Indian literature, such as *Susruta Samhita* (600 BC), classifies all 'substances' into *Sthavara* (immobile), e.g. plants, *Jangama* (mobile), e.g. animals. Parasara grouped angiosperms into *Dvimatruka* (dicotyledons) and *Ekamatruka* (monocotyledons). He characterised the former with *Jalikaparna* (reticulate-veined leaves) and the latter *maunlaparna* (parallel-veined leaves). For more details you may consult Chapter 1.

Greek scholars Hippocrates (460-377 BC) and Aristotle (384-322 BC) arranged animals into four major groups like insects, birds, fishes and whales. Theophrastus (370-285 BC), often referred to as 'Father of Botany', classified plants on the basis of their habit, form and texture into four categories: trees, shrubs, undershrubs and herbs. He gave names and descriptions of 480 plants in his book *Historia Plantarum*. In the late seventeenth century, another biologist John Ray, (1627-1705), based on extensive travel, described more than 18,000 plants and animals in his *Historia Generalis Plantarum*. He coined the term **species** for a group of morphologically similar organisms and also tried to differentiate between genus and species.

Another significant period for taxonomy or science of classification of organisms was that of Linnaeus. Carolus Linnaeus (1707-1778), a Swedish naturalist, often referred to as 'Father of taxonomy', published *Systema Naturae* (1735). His treatise, *Species Plantarum* (1753) contained description of about 6,000 species of plants and these were arranged according to his system of classification based on sexual characters. Linnaeus also introduced a system of nomenclature of plants and animals known as the **Binomial Nomenclature**. According to this method, the scientific name of a species consists of two parts — the first part represents genus to which the species belongs, while the second part represents the identity of the species to which the individual belongs. For example, humans are named *Homo sapiens* and the pea plants are called *Pisum sativum*.

In earlier systems of classification, only external morphological characters, observable with the help of unaided eye formed the sole basis of classification of plants and animals. The characters like root modifications, leaf venation and floral structures and number of cotyledons were used in classification of flowering plants. From Aristotle to Linnaeus, every one considered only limited features in the classification of organisms. As a result, the diverse living organisms in nature were placed into limited number of groups. This phase of taxonomy led to the development of artificial systems of classification. Later, the organisms were classified on the basis of natural affinities (i.e., the basic similarities in the morphology). This phase represents the classical taxonomy and constitutes the natural systems of classification.

An approach called **numerical taxonomy** or **phenetics** has emerged since 1950. As the name indicates, it employs numerical methods for the evaluation of similarities and differences between the species. In this method as many characteristics as possible are used for these comparisons, without extra emphasis being given to any one. Such comparison is possible due to the availability of computers. The organisation and analysis of data is core to this approach. More options can be processed with more data. All characters considered for analysis are given equal importance and weightage. This system of classification is considered better because it uses large number of comparable characters to assign a place to the species.

At the opposite end of the taxonomic spectrum are the biologists who assign taxonomic affinity based on evolutionary as well as genetic relationships amongst organisms besides morphology. They ignore the morphological similarity or difference. This system of classification is designated as **phylogenetic classification** or **cladistics** (*L. clados*:branch). Cladistics classifies organisms according to the historical order in which the evolutionary branches arose. This led to the emergence of **new systematics** (Sir Julian Huxley 1940) or **biosystematics**.

The improved tools and techniques for classification helped in the emergence of new branches of systematics, such as cytotaxonomy, chemotaxonomy or biochemical taxonomy and population systematics. These trends are based on the review of the earlier principles and the use of cytological and biochemical techniques, serology, molecular biology, comparative study of behaviour and of late, the use of computers

for evaluation of distinctions and similarities between the organisms.

Cytotaxonomy uses cytological information on cell, chromosomal number, structure and behaviour of chromosomes during meiosis for classification purposes. It has been found that number of chromosomes is constant in the species, for example man and potato have 46 and 48 chromosomes, respectively. Also, the size of chromosomes may vary, for instance, herbaceous plants have larger chromosomes than the woody plants. Information about the mode of pairing of chromosomes during meiosis is used by the taxonomists to understand the relationship between the species.

Chemotaxonomy is based on the chemical constituents of plants. The fragrance and taste vary from species to species. Ancient classifications for medicinal purposes were based on chemical information such as aroma in the case of spices. The chemical constituents of plant species have been found to be stable and do not change easily. The chemical information can be gathered from any part of the plant. The characters like presence of calcium oxalate crystals, called raphides, have been found to be common to 35 families. Chemical characters have also helped in establishing relationships and statistical evaluation of taxonomic information. The sequencing in DNA and chemical nature of proteins have also been used to establish similarities and affinities.

4.3 BASICS IN CLASSIFICATION

Classification means the ordering of organisms into groups. The branch of science that deals with the study of principles and procedures of biological classification is called Taxonomy. Taxonomy enfolds the following fundamental elements.

Nomenclature is assigning of names to organisms. It is the determination of correct name as per the established universal practices and rules. Every taxonomist has to follow these rules.

Classification deals with the mode of arranging organisms or group of organisms into categories according to a systematic plan or an order. The categories used in the classification of animals and plants are Kingdom, Phylum (Division in plants), Class, Order, Family, Genus and Species. Each category is a unit and can also be referred as **taxon** (pl. **taxa**)

Identification for taxonomic classification is carried out for an organism to determine its similarity with an already known organism. This implies assigning an organism to a particular taxonomic group. Suppose there are three plants say *a, b, c.* All represent different species. Another plant, say *d,* resembles *b.* The recognition of the plant *d* as identical to the already known plant *b* is its identification.

4.4 NOMENCLATURE

We give names to objects around us. You are also familiar with the common names given to pets and even domestic animals. Generally, names are in local or common language and are called vernacular names. Sometimes, group names like dog, cattle, tree or shrub are also given based on the similarities among the organisms.

Let us see how the names change with change of language. The plant Gurhal (Hindi) is also known as China rose (English), Padmacharini (Sanskrit), Jaba (Bengali), Mandara (Oriya), Jaswand (Marathi) and Dasavala (Kannada). It may have other names in other languages of our country. Similarly, the bird, Indian sparrow, is known as gauraiyya (Hindi), house sparrow (English), Pardal (Spanish) and Suzune (Japanese). The common name may include several organisms, such as 'Titlee' includes butterflies and moth. Sometimes the common names are misleading, for example, jellyfish and starfish are not fishes at all.

Scientific Names

It is not possible to identify the organisms universally based on their common names. Even biologists find it difficult. They must have a scientific name, which is acceptable all over the world. Such naming must be based on agreed principles and criteria. The scientific names ensure that only one name is given to an organism and description of the organism should help the other people to arrive at the same name in any part of the world. Each kind of organism, representing a species, is given a different name to distinguish it from others. One has to ensure that such a name has not been used earlier for any other organism. The following have been the practices of providing names to the organisms.

Polynomial Nomenclature

Before 1750, scholars who desired to designate a particular species usually used to add a series of descriptive words. Names became lengthy and difficult to remember. This can be illustrated with the example of *Caryophyllum*. The name given was *Caryophyllum saxatilis folis gramineus umbellatis corymbis* meaning *Caryophyllum* growing on rocks having grass like leaves and umbellate corymb arrangement of flowers. The resulting string of such Latin words was called a **polynomial system** of nomenclature.

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Binomial Nomenclature

The polynomial system was found cumbersome and often changed from scholar to scholar. In search of a better alternative, Carolus Linnaeus (1753) introduced a shorthand designation, binomial (L. *bi* : two; *nomen* : name), for each species. Ultimately, the **binomial system** became a common and established practice. Let us take the example of Mango and honeybee to understand the binomial system of naming. The scientific name given to mango is *Mangifera indica* which is latinised. *Mangifera* represents the genus and *indica* the specific epithet. Similarly, the honeybee is called *Apis mellifera*. Thus, you should remember now that every scientific name has a first word for *generic name* followed by a second word denoting *specific epithet*. Therefore, binomial nomenclature is the system of scientific naming of an organism using generic name as the first part and specific epithet as the second part.

Trinomial Nomenclature

Occasionally, three words are also used for naming the organisms, especially the animals. These include generic, specific and sub-specific parts. For example, the modern man is called *Homo sapiens sapiens*.

Any one can study, describe, identify and give a name to an organism provided certain universal rules are followed. These rules are framed and standardised by International Code of Botanical Nomenclature (ICBN) and International Code of Zoological Nomenclature (ICZN). The codes help in avoiding errors, duplication, confusion and ambiguity in scientific names. The codes are established and improved upon at International Botanical and Zoological Congress held from time to time. The names of bacteria and viruses are decided by International Code of Bacteriological Nomenclature and International Code of Viral Nomenclature. Similarly, there is a separate International Code of Nomenclature for Cultivated Plants.

Guidelines for Naming of Organisms

Let us study some of the universally accepted norms for nomenclature.

(i) A scientific name generally has two words in Latin or derived from Latin irrespective of their origin.

(ii) First word denotes the genus whereas the second one is for species.

(iii) Names are printed in italics or are separately underlined to indicate their Latin origin.

(iv) Generic name starts with a capital letter and the specific name with a small letter, e.g. *Mangifera indica*.

(v) The name of the author is written in abbreviated form after the species name and it is printed in Roman, e.g. *Mangifera indica* Linn.

(vi) Each taxonomic group can have only one correct name.

(vii) The name should be short, precise and easy to pronounce.

4.5 TAXONOMIC HIERARCHY

The main aim of a taxonomic study is to assign organism an appropriate place in a systematic framework of classification. This framework is called **taxonomic hierarchy** by which the taxonomic groups are arranged in definite order, from higher to lower categories. The categories used in the classification of animals are Kingdom, Phylum, Class, Order, Family, Genus and Species. However, in the case of plants, Division is used as a category in place of Phylum while remaining categories have same terminology. All the members of a taxon show similar characteristics, which are different from that of other taxa. Let us take an example of insects. It represents a group of organisms, which have common characteristics like three pairs of jointed legs by which they can be recognised and given the rank or category of the Class Insecta. The fishes, birds, mammals, algae, ferns, grasses are other examples of groups of organisms in classification. Both in animal

and plant kingdoms, the lowest category is the species and highest is the kingdom. The placement of group of individuals or organisms in species, genus and higher categories is determined by the similarities in their characters and the relationships. The categories in the hierarchy are thus in ascending order (Fig 4.1). As we go from the lowest rank species towards kingdom the number of similar characters decreases.

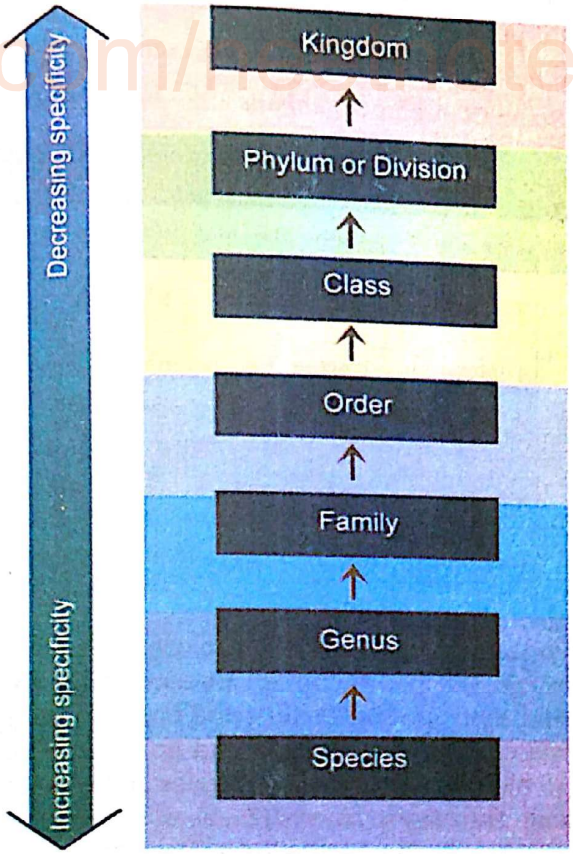

Fig. 4.1 *Hierarchy of taxonomic categories*

Species is a basic unit for understanding taxonomy as well as evolution. It is a group of individuals with similar morphological characters, which are able to breed among themselves and produce their own kind. You may recall the example of mango *(Mangifera indica)*, potato *(Solanum tuberosum)*, and lion *(Panthera leo)*. In this case, *indica, tuberosum* and *leo* are species of genera *Mangifera, Solanum* and *Panthera*, respectively. A genus may have more than one species as in *Panthera tigris* where *tigris* is another species. The individuals of species also represent population of species and they do not breed with individuals of other species.

Genus is a group of species which are related and share number of characters. For example, potato and brinjal though they constitute different species belong to the same genus *Solanum*. Similarly lion, leopard and tiger have several common features and are included in the genus *Panthera*. You will find that a genus may have a single species as in *sapiens* of genus *Homo* or it may have several species.

Family is represented by a group of related genera that are more similar to each other than with the genera of other families. The genera like *Solanum, Petunia, Atropa* based on the similarities are placed in the family Solanaceae. Animals such as lion, leopard, tiger from genus, *Panthera*, and cats from genus, *Felis*, are included in the family Felidae. However, if you observe and compare the features of a cat and a dog you will find distinct differences between them. They are placed in different families : Felidae and Cancidae, respectively.

Order is an assemblage of families resembling one another in a few characters. These characters are less similar as compared to many genera put in a family. Families like Solanaceae and Convolvulaceae are put in the order Polemoniales on the basis of some related floral characters. The families Felidae and Canidae of animals are included in the order Carnivora.

Class represents organisms of related orders. The order named Primata comprising monkey, gorilla and gibbon is put along with order Carnivora representing tiger, cat and dog. Both the orders are assigned to class Mammalia. There are other orders also in Mammalia.

Phylum includes all organisms belonging to different classes having a few common characters. Among the animals, the phylum Chordata includes classes Pisces, Amphibia, Reptilia, Aves and Mammalia, since all classes at some stage in their lives have common characters like presence of a notochord, dorsal hollow nervous system and pharynx perforated by gill slits.

Kingdom, in general, includes all organisms that share a set of distinguishing common characters. Plants are put in plant kingdom while animals are included in the animal kingdom. This is the highest category of classification.

The higher the category lesser will be the number of common characteristics of organisms

belonging to that category. For example, lion, tiger and leopard belonging to the genus *Panthera* have many common characteristics but differ widely from bat, elephant, gorilla and man although all of them belong to the class Mammalia. There may be other sub-ranks like sub-species below the species, sub-class between order and class, and sub-kingdom between phylum and kingdom.

Different categories to which human beings, dog, housefly, mango, wheat and tulsi belong are shown in Table 4.1 to give you an idea of the taxonomic hierarchy.

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Table 4.1 Taxonomic Categories of Some Common Organisms

Common Names	Scientific Names	Genera	Families	Orders	Classes	Phyla/ Divisions
Human	*Homo sapiens sapiens*	Homo	Hominidae	Primata	Mammalia	Chordata
Dog	*Canis familiaris*	Canis	Canidae	Carnivora	Mammalia	Chordata
Housefly	*Musca domestica*	Musca	Muscidae	Diptera	Insecta	Arthropoda
Mango	*Mangifera indica*	Mangifera	Anacardiaceae	Sapindales	Dicotyledons	Angiospermae
Wheat	*Triticum aestivum*	Triticum	Poaceae	Poales	Monocotyledons	Angiospermae
Tulsi	*Ocimum sanctum*	Ocimum	Lamiaceae	Lamiales	Dicotyledons	Angiospermae

4.6 SYSTEMS OF BIOLOGICAL CLASSIFICATION

The earliest systems of classification of organisms were simple and based on one or two characters. The criteria used for grouping were also very simple. For example, Aristotle and others grouped plants into trees, shrubs and herbs based on their habit. He also divided animals into two groups, one having red blood (Enaima) and the others without red blood (Anaima). The period from Greeks to middle of eighteenth century experienced crude methods of classification based on habit/habitat, and a few structural features. Sometimes, the resemblances among the organisms considered were superficial and were found to be unreliable. This resulted in arbitrariness and unreliable systems of classification. Let us study the various systems of classification proposed from time to time.

Artificial System

This system of classification was based on one or a few superficial similarities. Sometimes, it was even made a matter of convenience resulting in arbitrariness. No weightage was given to natural as well as the phylogenetic relationships. This was prevalent before and also during the period of Linnaeus. Even before him, Aristotle had also classified organisms on the basis of their habitat. The habitat was recognised as a criterion to group animals that live on land, in air, and in water. In this system quite diverse and unrelated animals were grouped together. For example, whale and fish were put in an aquatic group, bat and birds in one group. Another example of artificial system of classification of animals is to categorise them into two groups: (i) can fly (ii) cannot fly. One would easily put butterflies and bats in a group along with birds. You will find in the artificial system, the criteria considered for grouping are simple and easy to follow. However, it sometimes results in grouping together of quite heterogeneous and unrelated organisms in one group, while closely resembling ones are included in widely separated groups.

Linnaeus used number, union, length and certain other characters of stamens as the basis of classification of flowering plants. For example, he proposed classes as Monandria (1 stamen), Diandria (2 stamens) and Tri-and polyandria (3 and more stamens). Even this system has been labelled artificial since Linnaeus did not consider phylogenetic relationships and based his system on a few characters.

Natural System

Organisms, according to the natural system, are classified on the basis of natural affinities. It

also uses more number of characters rather than a single character for determining the similarities. Natural classification is mainly based on form relationships realising all information available at the time of collection of plant. The natural systems remained dominant before the idea of evolution was accepted. The most important and the last of the natural systems for classification of seed plants was proposed by George Bentham (1800-1884) and Joseph Dalton Hooker (1817 -1911). The characters taken for identifying similarities are homologous i.e. relationship of comparable structures in different organisms. (See Chapter 3)

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Phylogenetic System

This system is based on evolutionary sequence as well as the genetic relationships among the organisms. Darwin's book *On the Origin of Species by Means of Natural Selection* or *The Preservation of Races in the Struggle for Life* (1859) provided the support to taxonomy. The most important change was about the status of species from *fixity*, prevalent before Darwin, to a dynamic or ever changing one, dominant after Darwin. The evolutionary development of groups of organisms or even an organism from its origin to the present state, in addition to morphological characters, became the basis of classification of plants. Adolf Engler (1844-1930) and his associate Karl Prantl (1849-1893) published a phylogenetic system in the monograph *Die natürlichen Pflanzenfamilien*. They placed families and orders of the flowering plants in ascending series based on the complexity of floral morphology. The characters like one whorl of perianth or no perianth, unisexual flowers and pollination by wind were considered primitive as compared to perianth with two whorls, bisexual flowers and pollination by insects. The plant kingdom according to their classification, is further divided into divisions, sub-divisions, classes, orders and families. Asteraceae (sunflower family) among dicots and Orchidaceae (orchid family) among monocots are considered highly advanced. According to them, monocots are more primitive than dicots. This system considered evolution of angiosperms from a single source and the sequence of orders and families show parallel evolution.

4.7 CLASSIFICATION OF ORGANISMS

Two Kingdom Classification

The living organisms from the beginning have been grouped into plants and animals. This is also known as Two Kingdom System of classification. Linnaeus named these Kingdoms as Plantae (including all plants) and Animalia (including animals). Plants as you are familiar, are not mobile and have autotrophic mode of nutrition. They synthesise their food themselves. This is possible due to energy capturing mechanism in plants and the presence of green pigment. Animals on the other hand, are mobile and are not able to synthesise their food. They depend on plants for their food. According to this classification, trees, bushes, climbers, creepers, moss on a rock or a wall or a tree, green algae floating in a pond are put in plant kingdom. There are exceptions in characters like locomotion and mode of nutrition. For example, if plants are taken as organisms containing cellulose then tunicate group of animals would be considered plants. They have cellulose, branching pattern like plants, and they anchor to the bottom of the sea. It will also be difficult to distinguish unicellular organisms in a pond or a lake as plants or animals because of their green, brown or red colour. The Two Kingdom System groups together the organisms with true nucleus as eukaryotes and without true nucleus as prokaryotes. It also considers photosynthesising green algae and non-photosynthetic fungi as one group of plants. The Two Kingdom System takes unicellular and multicellular organisms together. Even unicellular organisms like bacteria were considered as plants. These are some of the shortcomings of this system. The placement of certain organisms into Two Kingdoms became difficult. The following examples further illustrate the drawbacks of the Two Kingdom System.

Mushrooms, bread moulds and other members of fungi were also included in plants though they are non-green and not able to synthesise their food. Fungi as a group of organisms differ from plants not only in lack of green colour but also mode of obtaining food. They have filamentous structures like green algae but differ from plants in composition of

cell wall. The inclusion of fungi in plant kingdom has become debatable.

Let us take the example of *Euglena*. It is green in colour; single-celled and moves like animals. Some of the taxonomists considered it a plant or plant-like and included it in the plant kingdom based mainly on the presence of chlorophyll. Others included it in animal kingdom along with the protozoans because of its locomotion by flagella. This also became a point of debate among the taxonomists.

You are familiar with bacteria, which are also unicellular organisms. The cell has nuclear material without well-defined nuclear membrane. It is a prokaryotic cell and resembles blue green algae. Thus, bacteria were included in the plant kingdom. However, they differ from other plants and animals due to the absence of a well-defined nuclear membrane and a distinct nucleus, as in an eukaryotic cell.

The Two Kingdom System of classification of organisms into plant and animals was found inadequate in the light of disputed position of organisms like fungi, bacteria and *Euglena*. The Two Kingdom practice also needed reconsideration due to the discovery of viruses and bacteriophages (another type of organisms), which can neither be considered as prokaryotes nor as eukaryotes. The viruses infect and cause diseases both in plants and animals. They lack cellular structure and depend on metabolic activity of the host. They have either DNA or RNA as their hereditary material. They, in fact, are considered on the borderline of living and non-living objects. Unlike, other organisms they survive in the living system only and are host specific. All the examples mentioned above necessitated the reconsideration of the two-kingdom system of classification of organisms.

Five Kingdom Classification

The debatable position of fungi, bacteria and viruses led to the replacement of Two Kingdom grouping by a Five Kingdom classification, which was proposed in 1969 by R.H. Whittaker. The Five Kingdoms are: Monera, Protista, Fungi, Plantae and Animalia (Fig. 4.2). The main criteria for classifying organisms into Five Kingdoms are complexity of cell structure, body organisation, the mode of nutrition, life style and the phylogenetic relationships. Observe Figure 4.3. You will find that evolution is reflected through increase in complexity of the cell (prokaryotic to eukaryotic), as well as in the organism (unicellular to multicellular). The mode of nutrition also diverged in the multicellular kingdoms *viz.* plantae, fungi and animalia. The ecological role of these three multicellular kingdoms was also established as producers, decomposers and consumers respectively. The organisms, according to the Five Kingdom System, are re-distributed in the additional three kingdoms while retaining the two kingdoms – Plantae and Animalia. All multicellular, mobile and heterotrophic organisms were assigned the Kingdom Animalia. The photosynthetic multi-cellular organisms were included in the Kingdom Plantae. Some unicellular plants like algae and protozoans were taken out from plant and animal kingdoms and were included in a separate Kingdom Protista. All bacteria and multicellular blue green algae with prokaryotic cells were transferred from plant kingdom to a new kingdom called Monera. Still the idea of separation of unicellular algae from multicellular algae does not find favour with many biologists and our growing knowledge about organisms may lead to a new system of classification.

Salient features of the five kingdoms are given below.

Kingdom Monera

This kingdom comprises single-celled prokaryotic organisms like bacteria and

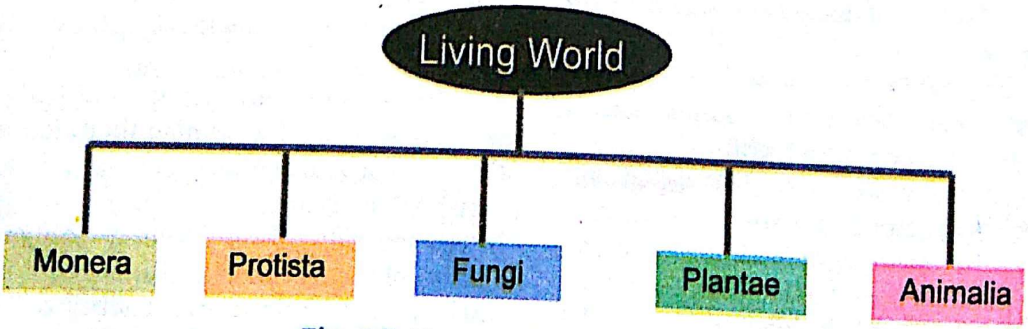

Fig. 4.2 *Five Kingdoms of organisms*

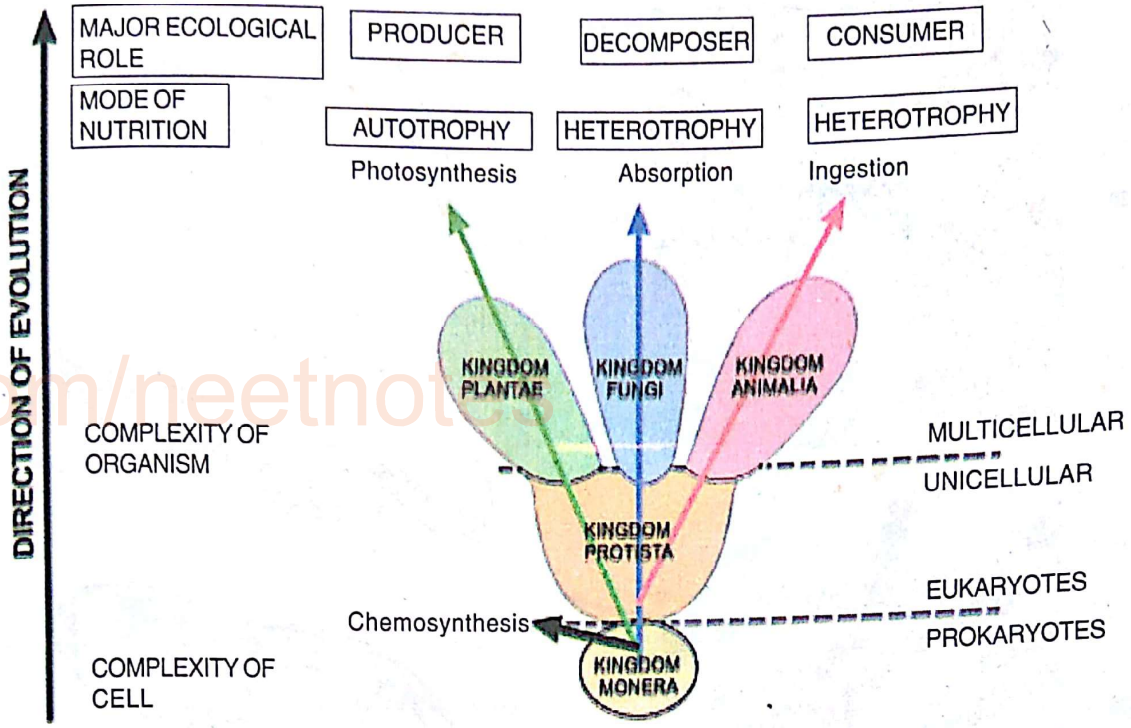

Fig. 4.3 *Five Kingdoms showing increasing complexity during evolution*

filamentous actinomycetes and photosynthetic blue green algae or cyanobacteria. The cyanobacteria are photosynthetic bacteria. However, they were called blue green algae also. The term algae was applied to the bacteria on the basis of their photosynthetic activity. Later on their structural relationships to bacteria were discovered with the help of electron microscope. They are appropriately referred as blue-green bacteria or cyanobacteria. The organisms representing this kingdom are microscopic forms. The nucleus and other organelles are without the enclosing membranes. The cells are prokaryotic with rigid cell walls. These microscopic monerian forms are considered most ancient and are found in places like deep ocean floor, hot deserts, hot springs and even inside the other organisms. Some forms are able to prepare their food with the help of light or chemical energy. Some members are heterotrophs and depend on their hosts.

Some of them are able to survive in extreme environments like high salt concentration, high temperatures and acidic/alkaline pH. They can also survive in anaerobic (oxygen free) conditions. These forms are grouped into true (Eubacteria) and the ancient bacteria (Archaebacteria). The latter are anaerobes (respire without oxygen) and live among the cattle's rumen and help in digestion of cellulose in the alimentary canal. They produce methane from carbon dioxide.

Bacteria are single-celled and have different shapes. They are rod-shaped (*Bacillus*, pl. *Bacilli*), spherical (*Coccus* pl. *Cocci*) or spiral (*Spirillum* pl. *Spirilli*). They reproduce by binary fission. You will study more about bacteria later in Chapter 9.

Bacteria are good decomposers and help in recycling of nutrients. They are also useful in making curd, and in fermentation. They cause diseases like diphtheria, pneumonia, tuberculosis and leprosy. They are also used in manufacture of anti-biotics to cure many diseases.

The blue-green algae also called cyano - bacteria in Monera include single-celled *Spirulina*, colonial *Nostoc* and filamentous *Oscillatoria* (Fig. 4.4). They are prokaryotic and photoautotrophic organisms.

Kingdom Protista

The members of the kingdom are unicellular and colonial forms usually found in aquatic habitats. The cells contain membrane-bound

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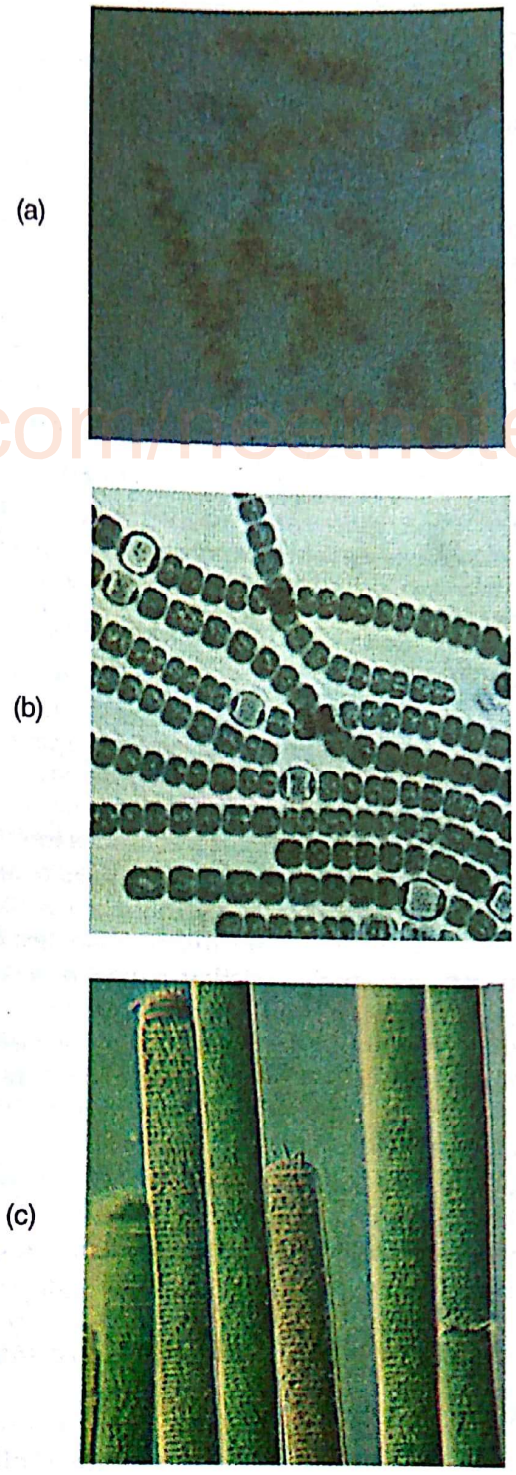

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Fig. 4.4 Monerans (a) *Spirulina* (b) *Nostoc* (c) *Oscillatoria*

nucleus (eukaryotic) and may have cilia or flagella for their movements (Fig 4.5). There are membrane bound cell organelles like nucleus, mitochondria, endoplasmic reticulum and the Golgi bodies. They have diverse forms including

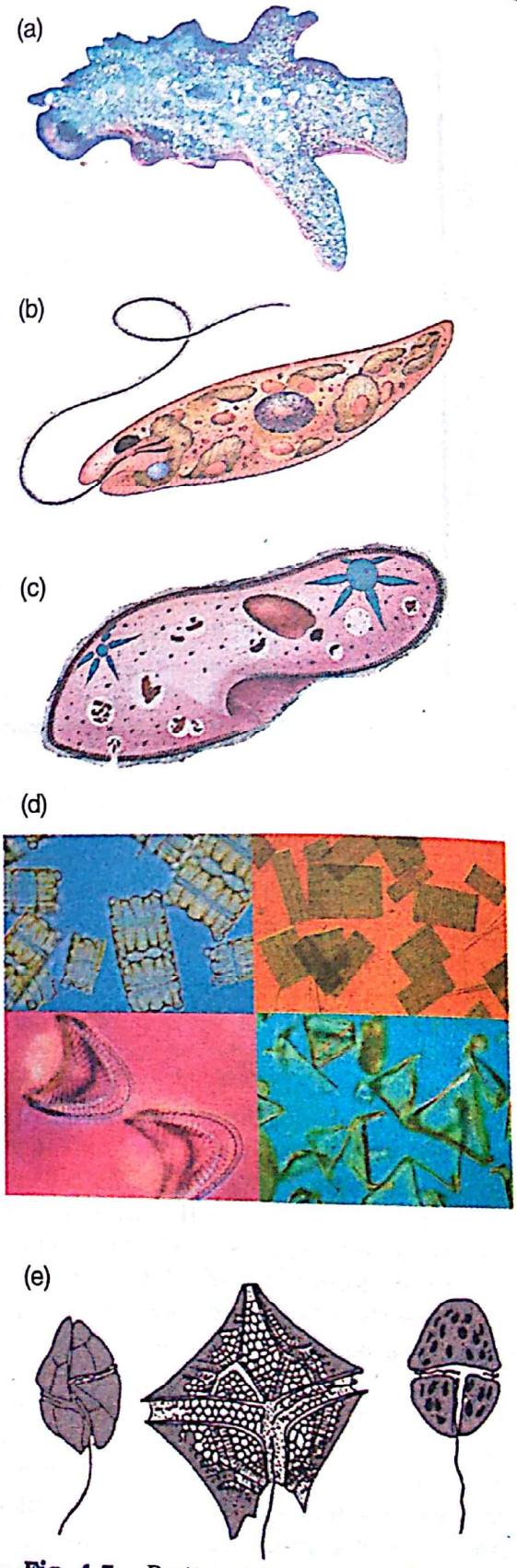

Fig. 4.5 Protists *(a) Amoeba (b) Euglena (c) Paramoecium (d)* Diatoms *(e)* Dinoflagellates

autotrophs, saprobes (living on dead matter) and even some members have parasitic mode of living. Those carrying out photosynthetic activity, such as algae and diatoms contain pigments. These microscopic forms are called phytoplanktons (*phyto*: plant, *plankton*: floating organisms). The reproduction in the members of this kingdom is through asexual division of haploid individuals as well as by a sexual process involving fusion of two cells and their nuclei.

On the basis of the mode of nutrition, the Protists are grouped as photosynthetic algae, decomposers (slime moulds), and predator protists (protozoans). Interestingly, some protists feed on other members of the kingdom. Protistan algae is represented by diatoms, dinoflagellates, flagellates like *Euglena*, and several other forms grouped into several divisions. These autotrophic protistans use light and carry out photosynthesis. In the absence of light they become heterotropic and ingest other protists like protozoa. Their dual mode of feeding resulted in their placement both in plant and animal kingdoms.

Saprobiotic members (slime moulds) occur both in cellular and acellular forms. They occur in damp places. The slimy mass of protoplasm with many nuclei (multi-nucleated) forms the body of myxomycete members. The thalloid body appears like amoeba and is called *Plasmodium*. It has structures like pseudopodia (*pseudo*: false, *podia*: feet) of amoeba, which help in movement. These are also used in engulfing food particles. During dry conditions whole body assumes a fruit like structure to overcome adverse conditions. Fragmentation helps in multiplication. The asexual spores called zoospores produced in sporangia are also means of reproduction. Interestingly, these spores also act as compatible gametes which fuse and form zygote. The thalloid body called plasmodium represents a diploid stage. They were included in plant kingdom in the Two Kingdom arrangement since they resemble fungi. Similarly, parasitic protozoans including *Amoeba* and malarial parasite were included in the phylum Protozoa of animal kingdom along with *Paramoecium*. The protozoans lack cell wall and hence directly ingest food. This mode of animal nutrition is called *holozoic*. Some forms like malarial parasite live as parasites in animals, including human beings.

Kingdom Fungi

Fungi grow as parasites on living organisms and as saprophytes on decaying organic matter in moist and warm conditions. Fungal forms are single-celled (Yeasts) as well as multicellular, complex, filamentous structures called mycelia (*sing. mycelium*) (Fig. 4.6). The mycelia are interwoven mass of thread-like hyphae (*sing. hypha*). The cells are eukaryotic and posses all cell organelles except the plastids. The cell wall is generally composed of chitin, a nitrogen containing carbohydrate, unlike plants where the cell wall is made up of cellulose. Some fungi also have cellulose in cell walls. They absorb soluble organic matter. The mode of obtaining food is called saprobiotic mode of nutrition and members are known as saprobes. Some are even parasites. They help in decomposition of organic matter such as dead bodies of plants and animals and play an important role in recycling of minerals.

The fragmentation of hypha helps in multiplication where each fragment grows into new mycelium. They re-produce by spores like zoospores, gonidia, conidia, ascospores and basidiospores. Sexual reproduction involves the fusion of similar (isogamy) and dissimilar gametes (oogamy). Fungi are further grouped, as given below, into classes based on the morphology of mycelium, mode of nutrition and mode of reproduction.

Phycomycetes are found in water on decaying organic matter and damp places and also as parasites. They have a multinucleated (coenocytic) and aseptate (without septa) mycelium. Asexual reproduction is through ciliated **(zoospores)** as well as non-ciliated spores called **aplanospores**. Both the kinds of spores are produced in sporangia. The sexual reproduction takes place by fusion of similar (isogamy) or dissimilar (oogamy) gametes. The common members are *Mucor* (Fig.4.6b), and *Albugo*.

Ascomycetes (*Ascus*: sac, *mycete*: fungus) are unicellular as well as multicellular fungi. In the latter, mycelium is septate. The asexual spores formed in chains are called conidia. The spores are formed exogenously, i.e. outside sporangium. They detach from the parent and form new mycelia. Sexual reproduction is through ascospores, which are formed endogenously (within the mycelium) in a sac like

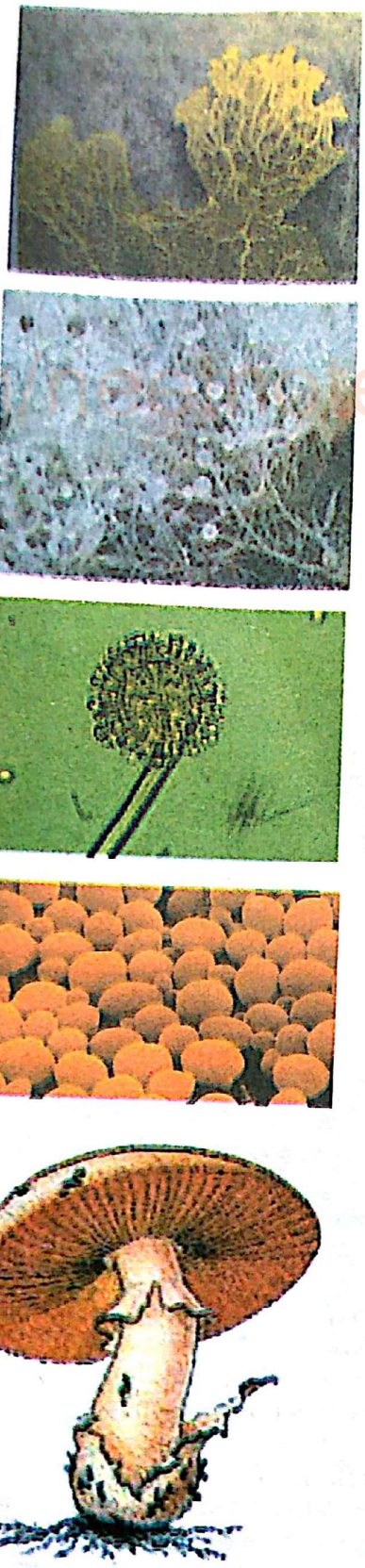

Fig. 4.6 Fungi *(a) Plasmodium (b)* Mucor *(c) Aspergillus (d)* Yeast *(e) Agaricus*

structure called **ascus** (pl. asci). The gametes involved in sexual reproduction are nonmotile compatible and are generally represented as + and –. Fusion occurs between them. The fusion of gametes is followed by reductional division that produces haploid ascospores. The fruiting body called **ascocarp**, is cup shaped and contains the spore bearing structures called asci (*sing. ascus*). Yeasts, *Penicillium*, *Aspergillus* and *Claviceps* are common examples.

Basidiomycetes are called club fungi because of a club shaped end of mycelium known as **basidium**. They have septate multinucleated mycelium. The sexual spores called basidiospores are generally four in number. They are produced outside the body (exogenous) unlike ascomycetes where they are endogenous. Two compatible nuclei fuse to form zygote, which undergoes meiosis and forms four basidiospores. The fruiting body containing basidia is a multicellular structure called **basidiocarp**. The common members are mushrooms (*Agaricus*), Smut *(Ustilago)* and Rust *(Puccinia)*.

Kingdom Plantae

This kingdom includes all plants, except some algae such as diatoms, dinoflagellates and other members of Monera, Protista and Fungi. These multicellular and photosynthetic forms are called green plants; they include red, brown and green algae, mosses, ferns, gymnosperms and flowering plants. The plant cells, unlike members of other kingdoms, have rigid cellulose cell wall, which prevents contraction. They also lack locomotion, which is a common feature of unicellular protists and animals.

You are already familiar with the process of photosynthesis by which the green plants synthesise their food by utilising water, minerals and carbon dioxide with the help of green pigment chlorophyll and solar energy. They are called photoautotrophs and primary producers in the ecological context. They provide food to consumers mainly the animals. The life, in fact, depends on plants for essential resources like food, oxygen and energy. Moreover, this kingdom also has forms with heterotrophic and parasitic modes of nutrition. Insectivorous plants, such as bladderwort and venus fly trap, consume insects. In general, the life cycle of this group of plants is completed in two phases – gametophytic and sporophytic, which alternate with one

another. The phenomenon is called alternation of generations. The kingdom is divided into several groups. You will study more about these groups in Chapter 5.

Kingdom Animalia

This kingdom comprises the animals, except the protozoans, which have been transferred to the kingdom Protista. The members of animalia are multicellular eukaroytes. The cells are without cell wall. They exhibit heterotrophic mode of nutrition, which is also called holozoic. They directly or indirectly depend on plants for their basic requirements particularly the food. The members have well-developed control and coordination mechanisms. Mostly they are free living, some are parasitic (live at the expense of others) and some live in association with others and lead a mutually beneficial life. This kingdom shows greater diversity as compared to the others. It is further sub-divided into Phyla based on morphology, physiology and ecological considerations. Salient features of various phyla are described in Chapter 6.

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Table 4.2 Some Important Characteristics of the Five Kingdoms

Kingdoms	Important Characteristics	Members
Monera	Unicellular, Prokaryotic, Autotrophic mode of nutrition, Asexual reproduction, Multiplication by amitosis	Archaebacteria (ancient bacteria), Eubacteria (true bacteria), Cyanobacteria or Blue green algae
Protista	Unicellular, Eukaryotic, Aquatic, Autotrophic and heterotrophic mode of nutrition, asexual reproduction by division into two, sexual reproduction by fusion.	Protozoans, Slime moulds
Fungi	Multicellular, Eukaroytic, No true tissues, Cell wall made up of chitin, Heterotrophic, saprobiotic, parasitic mode of nutrition (absorptive), Asexual and sexual reproduction by spores and gametes respectively.	Bread mould, Yeasts, Mushrooms
Plantae	Multicellular, non-motile forms, Eukaroytic, Cell wall made up of cellulose, Well developed tissues, Photosynthetic-autotrophic mode of nutrition, asexual reproduction by multiplication, distinct sex organs, Life-cycle exhibits alternation of generations	Algae, Liverworts, Mosses, Ferns, Conifers, Flowering plants
Animalia	Multicellular, motile forms, Eukaryotic, Well developed tissues, Heterotrophic mode of nutrition, Sexual reproduction with distinct sex organs, Control and coordination system, Distinct embryological development	Sponges, Cnidarians, Worms, Insects, Molluscs, Star fish, Fishes, Amphibians, Reptiles, Birds and Mammals

4.8 TAXONOMICAL AIDS

The laboratory and field studies are important for identification of various species. The information thus gathered about the species need to be stored for future studies. The actual specimens are also collected, preserved and stored for verification if required later on. This helps in identification of species and their placement in taxonomic hierarchy. There are several aids like herbaria and museums, which help in taxonomic studies. Let us study some of these aids.

Herbarium

A herbarium is defined as a collection of plants that usually have been dried, pressed and preserved on sheets. The sheets are arranged in accordance with any accepted system of classification. The storage of sheets forms a repository for future use. It provides quick refer-back system and is quite useful for people involved in taxonomic studies. The preparation of a herbarium involves several steps outlined below.

The collection of specimens requires regular field visits. We have to get information about the place, habitat, season and the time of collection. The field visit helps in gathering information about environmental conditions, human impact on the area and other general information from earlier records, maps or other sources. One has to carry certain simple tools for collection of specimens or their parts (Fig. 4.7). The individual parts of a plant are quite useful in identification. In the field, one requires digger for digging roots, scissor for cutting twigs, knife for woody twigs and a pole with a hook for

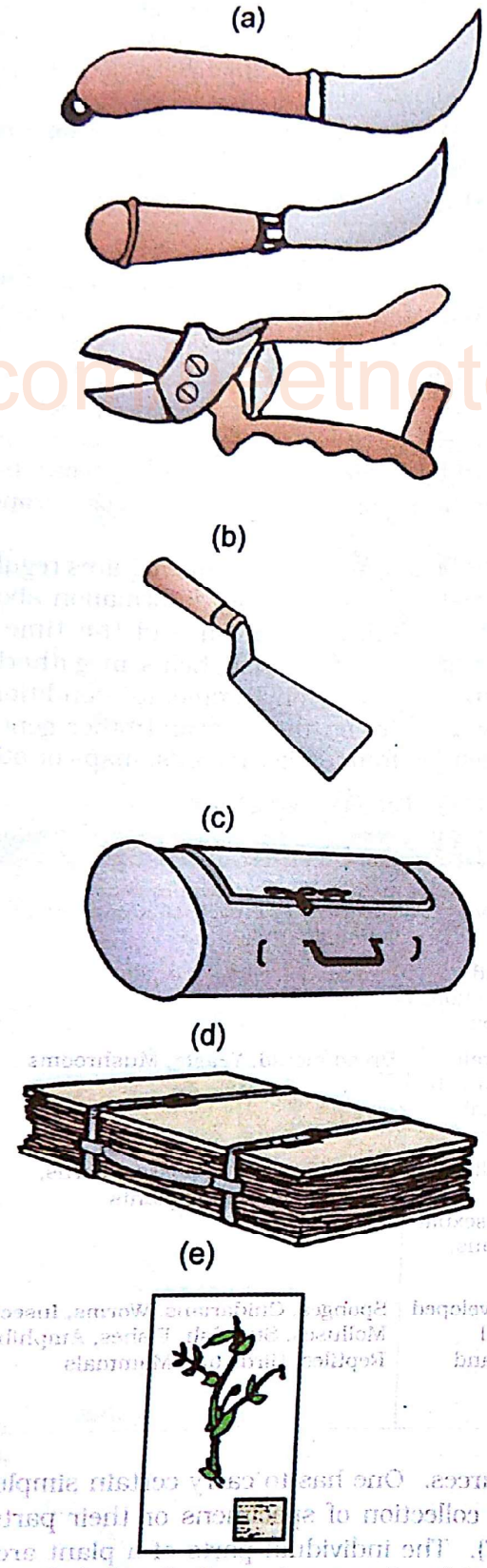

Fig. 4.7 Tools for collection and preservation of plants (a) Cutter (b) Digger-khurpi (c) Vasculum (d) Plant press (e) Harbarium sheet

collecting parts of tall trees. The collected specimens are carried in a box called vasculum to avoid loss of moisture and distortions by drying and shrivelling up. Polythene bags are also used for carrying the specimens. Taxonomy students carry a field notebook for recording every detail rather than depending upon the memory. The care is to be taken to ensure that in the case of herbs both vegetative and reproductive parts are there in the collected plant specimen. However, twigs with leaves, inflorescence and flowers are collected from the shrubs and trees. Generally sufficient number (5-6) of individual specimens of a particular kind are collected. This is necessary in case of damage during the process of transport and preservation. The specimens are referred to by the assigned collection numbers, which are called the field numbers.

The specimens are spread out, in shortest possible time. They are dried by keeping them between the folds of old newspapers. It is necessary to change these papers at regular intervals to avoid fungal growth. Complete specimens with all parts are dried in a plant press. The plant press consists of a set of two boards with straps, which help in tightening the newspaper sheets with specimens between the boards. The bigger specimens are not cut but they are folded generally in *n* or *w* form. Some leaves are spread for showing dorsal surface and others to show ventral surface. The dried specimens are pasted on the herbarium sheets of standard size (29×41 cm). Specimens before fixing are kept upside down on paper sheets, glue or adhesive is applied and they are mounted on the herbarium sheets. All the pasted specimens are sprayed with fungicides like 0.1% solution of mercuric chloride, pesticides like DDT, naphthalene and carbon disulphide to check the growth of fungus. The heavy parts of plant like seeds, fruits are put in a packet and attached to the sheet.

It is necessary to label each herbarium sheet. It should carry the information about place/ locality, name of collector, date and time, common english name, vernacular name and scientific name of the species along with author's name. The name of the family and genus are also given in the case of already identified species.

The sheets are arranged according to a particular system of classification and stored preferably in metallic cupboards (Fig. 4.8). The area is disinfected to control insects and other

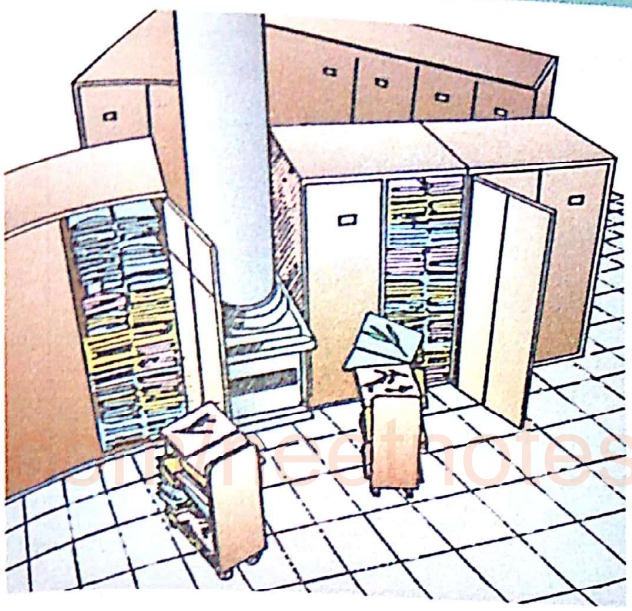

Fig. 4.8 *Herbarium showing stored specimens*

pests and humidity is monitored to check fungal growth. The information about the specimens is compiled and published in a form of a book (Flora), which gives a list of total plant species in a particular region, country or continent together with a brief description. We have several floras in India such as Flora of British India, Flora of Delhi, Flora Simlensis and others.

Botanical Gardens

These are collections of living plants maintained for reference. The plant species grown are used for identification and classification. It is considered natural and economical reference system. If you visit the Indian Botanic Garden, Howrah, National Botanical Research Institute (NBRI), Lucknow and other botanical gardens you will find each plant in the garden with scientific name and family of the plant mentioned on the labels.

Museums

Museums have collection of preserved plants and animals for study and reference. The collection of specimens helps in gathering the first hand information about the habitat, soil and organisms of the area. The museum is prepared to preserve plants like algae, fungi, mosses, and ferns, parts of gymnosperms since they cannot be kept in the herbaria. The animals are also preserved in the museums. The specimens are fixed in chemical solutions and are preserved for longer duration. The specimens are correctly identified and labelled. They are stored and a catalogue is prepared for future reference. The objective of preparing a museum is to record information and preserve specimens for taxonomic studies. It does not intend to kill or destroy the animals unnecessarily. These days collection of animals by school and undergraduate students are not encouraged. Being student of biology, you may like to collect and preserve plants, plant parts and dead animals like snakes, fishes, molluscs, insects and others. One should also appreciate the need of getting first-hand information about the organisms to study their characters, identification, naming and classification for better understanding of the organisms.

Zoological Parks

These help to know about wild animals, their food habits, behaviour, etc. People especially the children visiting zoos are excited and enjoy the visit by seeing a variety of animals (the animals are provided with conditions as close as possible to the natural habitats). A scientific purpose of the zoo is to breed the animals which otherwise are facing a threat in their natural habitat. You may be aware that animals are facing poaching and habitat destruction due to developmental activities. Information about common name and scientific name is also displayed in the zoological park.

Keys

A scheme for identification of plants and animals is known as a **Key**. Taxonomic keys are based on the contrasting characters. Separate taxonomic keys are required for each taxonomic category like family, genus or species. These are more useful in identification of unknown organisms. Being analytical in nature these are generally of two types: (a) Yoked or Indented and (b) Bracketed.

The **indented key** provides sequence of choices between two or more statements of characters of species. The user has to make correct choice for identification. We can identify the genera in the family Ranunculaceae by using the following indented key by considering the characters of the carpel and fruit of the specimens. The first choice starts with carpel with single ovule and achene type of fruit in contrast to carpels being many ovuled and fruits being follicles.

Indented Key

Character	Genus
Carpel single-ovuled, fruit achene	
Leaves opposite, compound	
petals absent, leaves without tendril	*Clematis*
Petals present	
Third or terminal	
leaflet modified into tendril	*Naravelia*
Leaves alternate or radical	*Anemone*
Carpel many-ovuled; fruit follicle	
Carpels united at base; flowers regular	*Nigella*
Carpels free at the base; flowers irregular	*Aconitum*

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Bracketed Key

Character	Genus
(1) Carpel single-ovuled; fruit achene _____	2
(1) Carpel many-ovuled; fruit follicle_____	4
(2) Leaves opposite; compound_____	3
(2) Leaves alternate, radical_____	*Anemone*
(3) Petals absent, leaves without tendril_____	*Clematis*
(3) Sepals as many as petals; third or terminal leaf modified into tendril	*Naravelia*
(4) Carpels united at the base; flowers regular _____	*Nigella*
(4) Carpels free; flowers irregular _____	*Aconitum*

*In the **bracketed key** the pairs of contrasting statements are used for identification. The number on the right indicates the next choice of paired contrasting statements.*

Keys are also used for identification of animals. Let us try to identify fish, frog, snake, bird, bat and cat using the given key. Distinguishing features are recorded for each group. Questions are framed to get only one answer. It could be 'Yes' or 'No'.

1. Does it have external ears ?	Yes	go to 2
	No	go to 3
2. Does it fly ?	Yes	Bat
	No	Cat
3. Does it fly ?	Yes	Bird
	No	go to 4
4. Does it have limbs ?	Yes	Frog
	No	Go to 5
5. Does it have gills ?	Yes	Fish
	No	Snake

Other taxonomic aids are monographs, which give comprehensive account of complete compilation of available information of any one family or genus at a given time. Manuals contain compiled information about area covered, keys, description of families, genus and species. Publications like periodicals and dictionaries are brought out to provide information about new additions and updated information.

SUMMARY

There are millions of living organisms with a vast variety of shape, size and form. They are found in diverse habitats. It is difficult to remember and describe each and every organism. Systematics is a branch of science dealing with identification, naming and classification of organisms. It is based on the study of characters as well as the relationships in evolutionary context. Taxonomy also connotes the same functions. Both the terms are synonyms. The characters used for taxonomical studies are morphological, cytological, physiological and ecological including the behavioural aspects. Systematics is useful not only for biologists but also in medicine, agriculture, forestry and industry.

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The earlier systems of classification were based on one or a few characters. Gradually natural affinities became the basis of classification. After the publication of Darwin's theory of evolution, the classifications began to be based on phylogentic relationships. Now information about cell structure, chromosomes, and chemical constituents are being used to establish the relationships. The use of computers and numerical methods for analysis and evaluation of information about the organisms helps in establishing evolutionary relationships on the basis of large number of characters.

Plants and animals are given names for identification. The practice of use of common names in local language creates confusion. Hence taxonomists started assigning universally acceptable scientific names to organisms. According to the binomial system of nomenclature, the name of an organism consists of two words, the first representing the genus and the second the species. In the two kingdom system of classification organisms are put either in plant or animal kingdom. In the Five Kingdom Classification System, the Kingdom Monera comprises unicellular bacteria, cyanobacteria and blue green algae. Some of these can survive in extreme environments and many play role of decomposers and help in recycling of nutrients. The members of kingdom protista are single-celled organisms generally found in aquatic habitats. They include autotrophs, saprobes and parasites. Kingdom fungi has non-green, unicellular and multicellular forms with eukaryotic cells. There are filamentous forms with saprobiotic mode of nutrition. They act as decomposers and help in recycling of minerals. Kingdom Plantae includes all multicellular, photosynthetic eukaryotic forms. They have well-established mechanisms for absorption and conduction of water, minerals and solutes.

Animalia forms the fifth kingdom and is represented by multicellular, eukaryotic forms with heterotrophic mode of nutrition. Some lead parasitic life style still others have adopted mutually beneficial mode of living. All are complex forms with well-organised organ-systems.

Herbaria, botanical gardens, museums and zoological parks are repositories of information useful for taxonomical studies. Herbaria are permanent records of plant specimens, which are collected, preserved, identified and classified. Botanical gardens are repository of living plants used mainly for taxonomic studies. The museums and zoos also help in the study of animals. Keys are the devices used for identification. They are based on contrasting characters.

EXERCISES

1. How is systematics relevant to other branches of biology?
2. Explain the utility of systematics.
3. Distinguish between cytotaxonomy and chemotaxonomy.
4. Numerical taxonomy is likely to furnish more reliable and stable information on the relationships of taxa for classification. Discuss.

5. Explain the role of chemical information in classification of plants.
6. Name the characters considered in cytotaxonomic studies.
7. Write the names of two important books brought out by Linnaeus.
8. Why was Linnaeus' system of classification considered an artificial system?
9. Give the main differences between natural and phylogenetic systems of classification.
10. What are the advantages of giving scientific names to the organisms?
11. Binomial nomenclature is the most acceptable mode of naming organisms. Why?
12. How do the biologists arrive at the universally acceptable names of plants and animals? Discuss.
13. What were the reasons for discarding the idea of fixity of species?
14. What is the correct way of writing a scientific name? Illustrate with example.
15. Why scientific names are written in italics?
16. Define taxonomic hierarchy? List the categories used in classification of organisms.
17. Define the terms species, genus and taxon.
18. What are the inadequacies of Two Kingdom System of Classification?
19. Compare the salient features of Monera with Protista.
20. Describe the important characters of kingdom Plantae.
21. Explain the distinguishing features of Animalia.
22. What do you understand by herbarium? Explain the various steps involved in preparing a herbarium specimen.
23. Botanical gardens are living herbaria? Comment.
24. What is the role of keys in taxonomy? Illustrate with example.

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Chapter 5

CLASSIFICATION OF PLANTS

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Plants, as discussed in the previous chapter, were frequently classified on the basis of their use, form and structure. Earlier systems classified them on the basis of their habit, as trees, shrubs, undershrubs and herbs. Gradually, the natural affinities in addition to morphological features became the major consideration for grouping plants. Studies on evolution helped in understanding the phylogeny of organisms. Taxonomists started using the phylogenetic relationships for classification purposes. This chapter deals with the classification of plants into major groups and their important characteristics. These major groups constitute the plant kingdom or Kingdom Plantae.

5.1 CLASSIFICATION OF PLANTS

In majority of systems which recognised only two kingdoms of organisms, the plants were included in the Plant Kingdom, whereas the animals were placed in the Animal Kingdom. For the classification of plants several systems were proposed from time to time. While using the number and position of stamens, Linnaeus divided flowering plants into 23 classes starting with the class Monandria with a single stamen ending with polygamia. He included all non-flowering plants such as algae, fungi, lichens, mosses and ferns in 24th class called Cryptogamia. His system was labelled as artificial system since it was based on a few characters. He, himself stated that his system of classification did not consider the relationships. Moreover, his system places widely unrelated families of monocotyledons and dicotyledons in one class.

Subsequently, several systems of classification of plants were proposed by taxonomists. It is not possible to discuss all of them. We will describe a common system which divides the plant kingdom into two sub-Kingdoms, named as Phanerogamae and Cryptogamae, considering the presence or absence of flowers and seeds (Fig 5.1). All plants which bear seeds are included in Phanerogamae (*Phaneros* : visible; *gamos* : marriage), whereas the Cryptogamae (*Kryptos* : concealed; *gamos* : marriage) covers all non-flowering plants such as algae, fungi, lichens, mosses and ferns. The Cryptogams are further classified into Divisions - Thallophyta, Bryophyta and Pteridophyta, each having classes assigned on the basis of similarities and differences among each group. The Phanerogams are also known as spermatophytes (*sperma* : seed; *phyton* : plant) since they produce seeds. These seed bearing plants are further grouped into two Divisions – Gymnospermae and Angiospermae. The gymnosperms (*gymno* : naked; *sperma*: seed) are represented by cycads, pines and cedars. These have naked ovules or seeds without any covering. The angiosperms include all the flowering plants which produce seeds and have ovules enclosed in an ovary or fruit. Another important characteristic used for distinguishing phanerogams from the cryptogams is the presence of well-developed vascular tissues for conduction of water, minerals (xylem) and food (phloem), respectively. The plants having vascular tissues (Pteridophytes, Gymnosperms and Angiosperms) are also called Tracheophytes (*Trachea* : wind pipe; *phyton* : plant).

You have learnt the distinguishing features of five kingdoms in the previous chapter. Here we will discuss the major plant groups which were included in the plant kingdom and later assigned to kingdom Plantae of the Five Kingdom

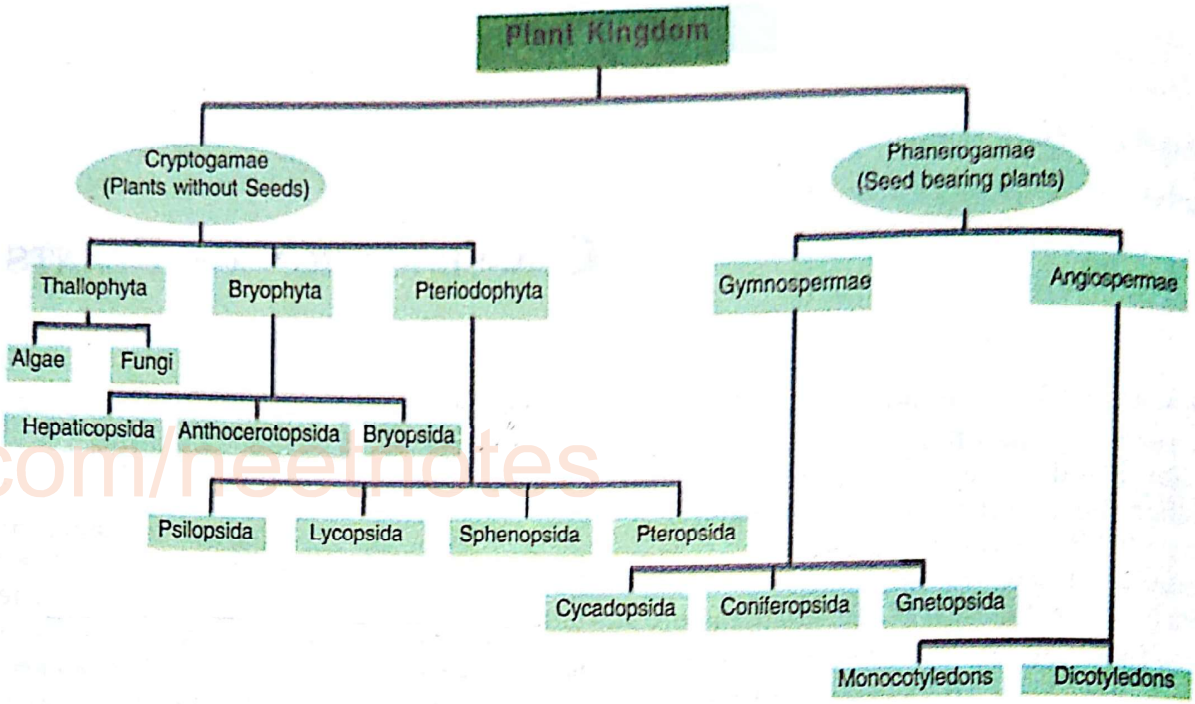

Fig. 5.1 *Classification of plants*

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System. The groups described here include algae (other than blue-green), bryophytes, pteridophytes, gymnosperms, and angiosperms. Fungi is also excluded since it has been given the status of a separate Kingdom discussed in chapter 4. Only the general characters of these groups along with the basis of their further classification are given. This will be followed by the classification system for flowering plants (angiosperms).

5.2 THALLOPHYTA – ALGAE

Algae and fungi are considered together in Thallophyta, though there is a basic difference in their mode of nutrition. Algae are autotrophic whereas fungi exhibit heterotrophic nutrition. Algae occur in a variety of habitats such as water, land as well as on the other plants and even animals. Some grow in marine water and are called seaweeds. The thallus (Fig. 5.2) is unicellular flagellated (*Chlamydomonas*) or non-flagellated (*Chlorella*), a colonial form (*Volvox*) or may even be of the filamentous type (*Ulothrix* and *Spirogyra*). In some forms, the thallus is flattened and leaf-like (*Laminaria*) and anchors to the rocks with the help of hold-fast. The unicellular protist algae are included in this figure for comparison. A variety of pigments in algae provide different colours. The green algae have mainly chlorophyll *a* and *b* along with carotenoids as photosynthetic pigments. The chloroplasts containing green pigment are of different shapes like girdle and spiral. The other pigments like fucoxanthin (brown), phycoerythrin (red) and phycocyanin (blue) provide colour to the algal forms. The red algae may secrete and deposit calcium carbonate and appear like corals. Common red algae are *Polysiphonia*, *Batrachospermum* and *Gelidium*.

Algae reproduce vegetatively by fragmentation (*Sargassum*), budding or tuber formation. A variety of motile and non-motile spores are involved in their asexual reproduction. The life-cycle has distinct haploid and diploid phases exhibiting phenomenon of **alternation of generations**. Based on the pigment colour, nature of stored food material and cell organisation, algae are further sub-divided into green (Chlorophyceae), brown (Phaeophyceae) and red (Rhodophyceae), etc.

5.3 BRYOPHYTA

The group derives its name from the mosses (*Gk. Bryon*: moss; *phyton*: plant) which grow on the rocks, walls and tree trunks in moist and shady places. You might have observed the moss growing as a green mat on the damp walls, during and after the rainy season.

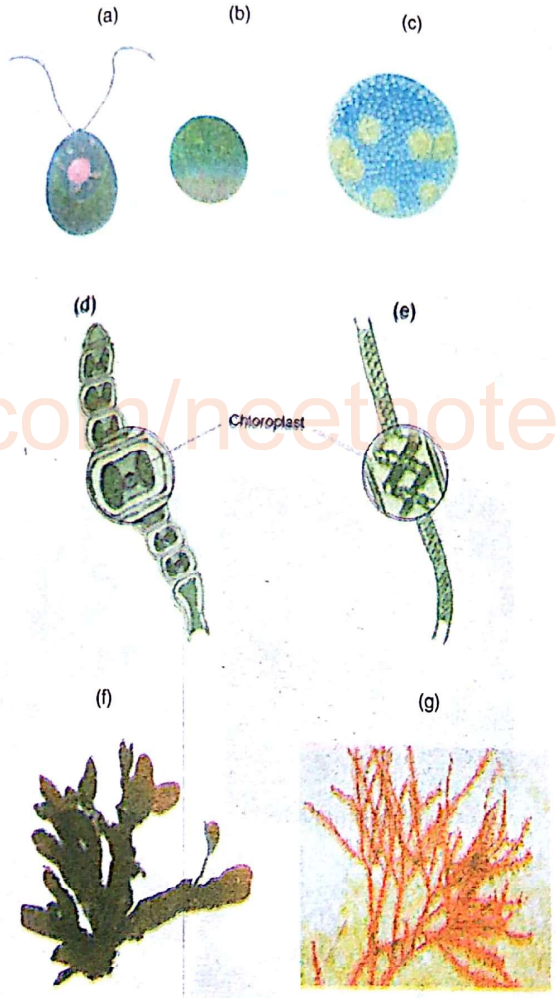

Fig. 5.2 *Algae : (a) Chlamydomonas (b) Chlorella (c) Volvox (d) Ulothrix (Girdle shaped chloroplast) (e) Spirogyra (Spiral shaped chloroplast) (f) Laminaria (g) Gelidium*

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The thalloid body is either flat (*Riccia*) or liver-shaped (*Marchantia*), hence these are also known as **Liverworts** (Fig. 5.3). In some forms such as moss the plant body is differentiated into stem- and leaf-like structures. The leaves are spirally arranged on stem-like appendages. The thallus is attached to the substratum - soil, rocks, walls or tree barks, with the help of root-like structures called **rhizoids** which help in the absorption of water. The thallus may also directly absorb moisture from the air. In contrast to the pteridophytes and seed-bearing plants, the plant body in bryophytes lacks vascular tissues. Mosses also retain moisture like the sponges. You might have observed dried moss being used for gardening purposes. Moss covered with fine net along with a rod is used as a moss stick for growing ornamental plants. The members reproduce both vegetatively through fragmentation, gemmae and tubers, as well as by sexual methods. The sexual reproduction involves male (antheridium) and female (archegonium) sex organs embedded in thallus and at the apex of leafy branches. Water is essential for fertilisation. The plant body in bryophytes represents the **gametophytic** phase and bears haploid gametes in the gametangia. The zygote formed by the fusion of male and female gametes develops into sporophyte which represents the sporophytic diploid phase and remains attached to the haploid gametophyte. The spores are produced in the sporophyte after meiosis or reduction division. The spores have 'n' chromosomes representing the haploid phase

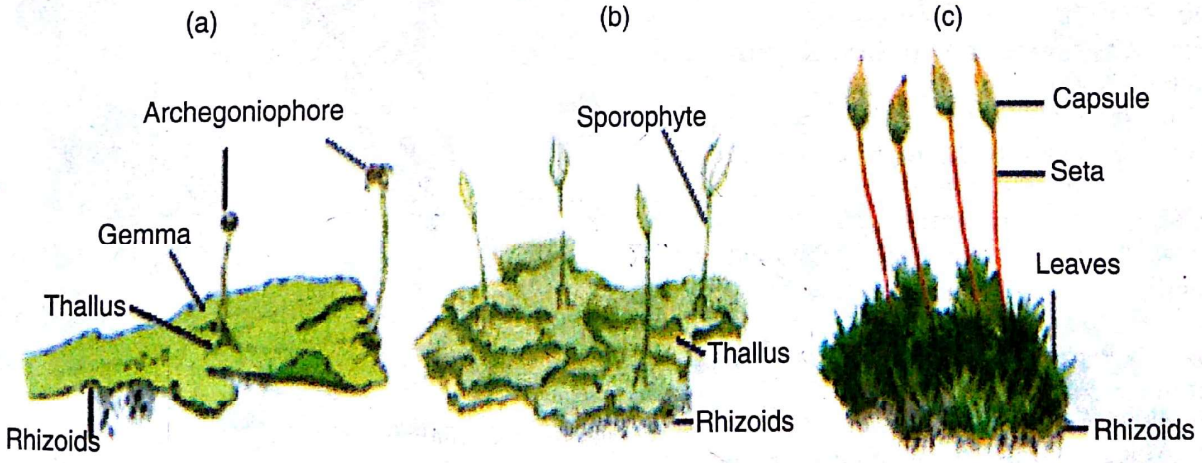

Fig. 5.3 *Common bryophytes (a) Marchantia (b) Anthoceros (c) Funaria*

and germinate to form the gametophyte. These two phases – gametophytic and sporophytic – alternate with one another. This phenomenon is called alternation of generations. Based on the structure of plant body and the method of sexual reproduction, bryophytes are divided into three classes – Hepaticopsida, Anthocerotopsida and Bryopsida. These are represented by *Marchantia*, *Anthoceros* and *Funaria*, respectively (Fig. 5.3 a,b,c).

5.4 PTERIDOPHYTA

This group of plants derives its name from the fern, *Pteris* which also represents salient features (*Pterido* : Pteris; *Phyton* : plant) of group. They occur in humid and tropical climates and usually grow on soil, rocks, in ponds and as epiphytes on other plants. These plants are also raised in pots as ornamentals. Amongst them, the tree ferns grow up to a considerable height appearing like small palm tree. Some members like *Equisetum* grow in the vicinity of water bodies. The most common members of this group are ferns *(Dryopteris, Pteris)*, *Lycopodium*, *Selaginella* and *Equisetum* (Fig. 5.4). A typical pteridophyte as represented by fern has a plant body differentiated into distinct underground stem-like rhizome, bearing roots, and aerial shoot with leaves. It helps the plant to regenerate if the aerial parts are destroyed by fire or animals. The pteridophytes have primitive vascular system. *Adiantum*, yet another well-known fern, shows a characteristic mode of multiplication. Whenever its leaf tip comes in contact with soil it develops adventitious roots and forms a new plant. As such it is also called the **walking fern**. You might have observed a similar mechanism of vegetative propagation through runners and suckers.

The leaves are of two types. Some ferns have simple leaves with a single vein whereas the others have compound leaves, comprising several leaflets (pinnules) like the flowering plants. The compound leaves in *Adiantum* with black shining petiole are beautifully arranged to provide appearance of a maiden's hair (Fig.5.4), hence it is also given the name **maiden hair fern**. Sporangia are borne on the ventral surface of the leaves as yellow or brown spots or sori (sing. sorus), arranged like pearls. These spore-bearing leaves are called **sporophylls**. The sori bear groups of sporangia (*sing. sporangium*)

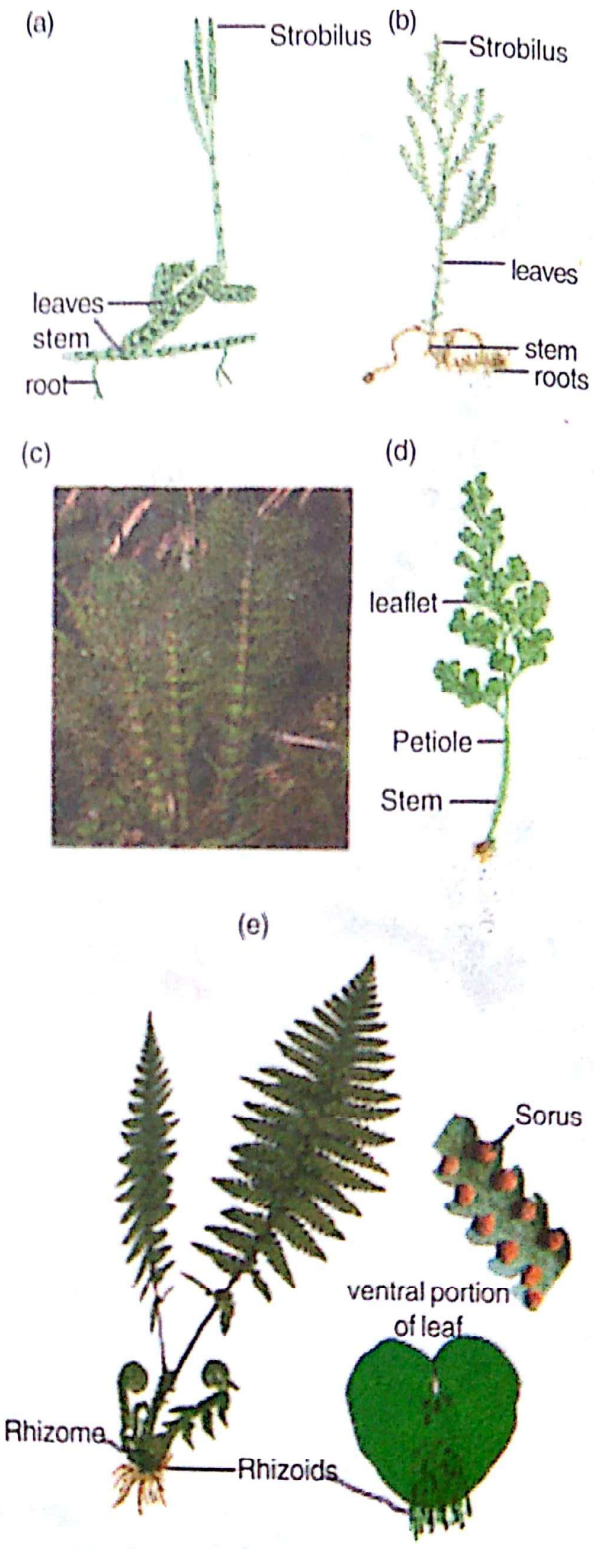

Fig. 5.4 *Pteridophytes : (a) Lycopodium (b) Selaginella (c) Equisetum (d) Adiantum (e) Pteridium and its prothallus*

full of spores. The plant body represents sporophyte and is diploid (2n). The spores formed after meiosis are haploid (1n) and are dispersed through a special mechanism present in the sporangium. These germinate on moist soil to form a thallus-like structure, known as **prothallus** which is multicellular and represents the gametophytic phase. The male (antheridium) and female (archegonium) sex organs develop on the ventral surface of the prothallus. The sporophytic (diploid) and gametophytic (haploid) phases alternate with one another and thus alternation of generations is fully perceivable. The dominant phase is sporophytic unlike the gametophyte in bryophytes. The Pteridophyta is divided into four classes, viz. Psilopsida, Lycopsida, Sphenopsida and Pteropsida, on the basis of organisation of plant body including the nature of leaf, vascular system, and location of sporangia.

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5.5 GYMNOSPERMS

Gymnosperms are vascular plants with naked seeds and are represented by conifers, which grow in cool climate of hills, sometimes using melting snow as a source of water. However, some like cycads and members of Gnetales thrive in warm climate. We may find a few members looking like palms growing in the plains. The most common one is *Cycas*. People in plains have started growing conifers like *Araucaria* as ornamental evergreen plants. The common gymnosperms are *Abies*, *Cedrus*, *Pinus*, and other timber-yielding species (Fig. 5.5). These are woody trees and only a few are bushy trailing or climbing shrubs like *Ephedra* and *Gnetum* with distinct root, stem and leaves. They grow successfully because they have a well developed vascular system with xylem without vessels and phloem without companion cells. These help them to absorb and conduct water in tall trees. They, being without flowers, have also acquired seed habit which enables them to overcome the requirement of external water for transfer of male gamete to the female gamete. The sporangia formed on special leaf-like structures called **sporophylls** are of two types. The megasporophyll (*mega*: big; *sporophyll*: Leaf-like structures with sporangium) bears megasporangium (ovule). The microsporangium, also called pollen sac, is enclosed in a microsporophyll (*micro*: small; *sporophyll* :leaf-like structures). The microsporophyll and megasporophyll form the male and female cones, respectively, which are so characteristic of conifers. The microsporangium contains pollen grains which are transferred by wind to megasporangium or the ovule. These produce male and female gametes, respectively.

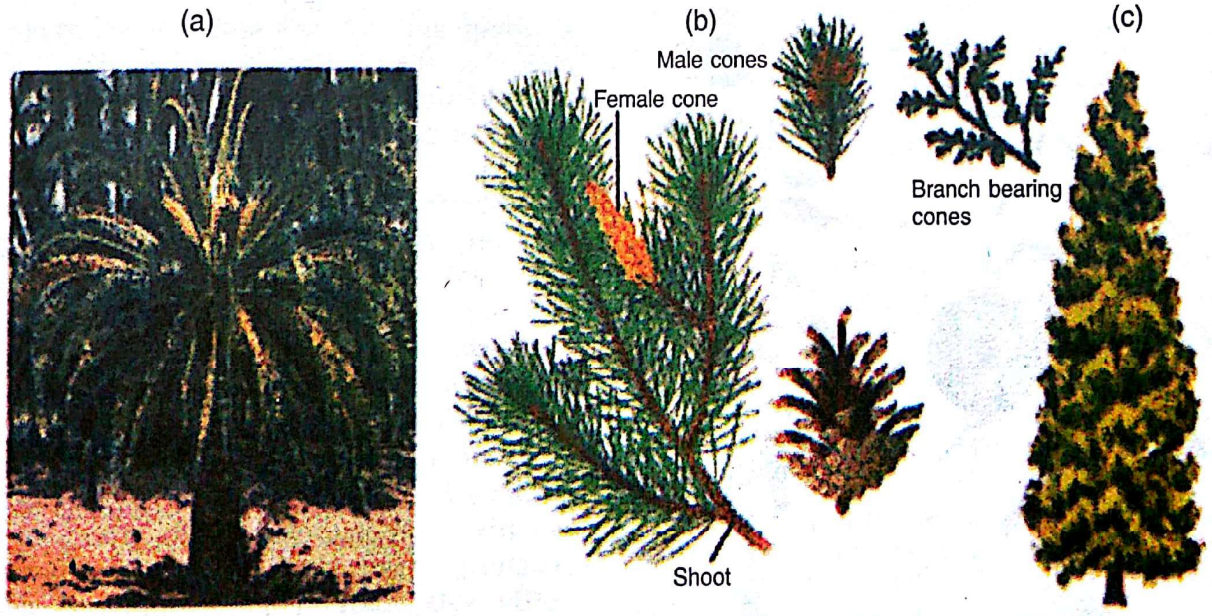

Fig. 5.5 *Gymnosperms : (a) Cycas (b) Pinus (c) Cedrus*

A diploid zygote formed after fertilisation of the egg develops into embryo within the ovule and the ovule ripens into a seed. However, in *Cycas* the megasporophylls are not organised in definite cones. The gymnosperms show a distinct alternation of generations where the dominant phase is represented by the sporophyte. These are further divided into three classes – Cycadopsida, Coniferopsida and Gnetopsida on the basis of nature of leaves, wood, vascular system and reproductive structures.

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5.6 ANGIOSPERMS

The angiosperms are seed-bearing plants well adapted to the terrestrial life and they occur in diverse habitats like cold tundra to hot tropical and even desert areas. They also thrive well in aquatic habitats. These plants represented by trees, shrubs and herbs are either monocotyledons or dicotyledons (Fig. 5.6). They have a body well differentiated into root, stem and leaves. They unlike Gymnosperms have a highly developed vascular tissue system with vessels in the xylem and companion cells in the phloem. Flowers in the angiosperms represent specialised sporophylls found in gymnosperms where the stamens can be equated with microsporophylls, carpels with the megasporophylls. The phenomenon of pollination, i.e. transfer of pollens from stamens to carpels, is an unique mechanism in angiosperms. It is facilitated by wind, water, insects, birds and other animals including man. After fertilisation, the ovules remain enclosed in the ovary, a character which differentiates them from gymnosperms. The seeds, in angiosperms thus remain enclosed in the ovary which ultimately develops into a fruit. These are dominant green flowering plants of the present day vegetation.

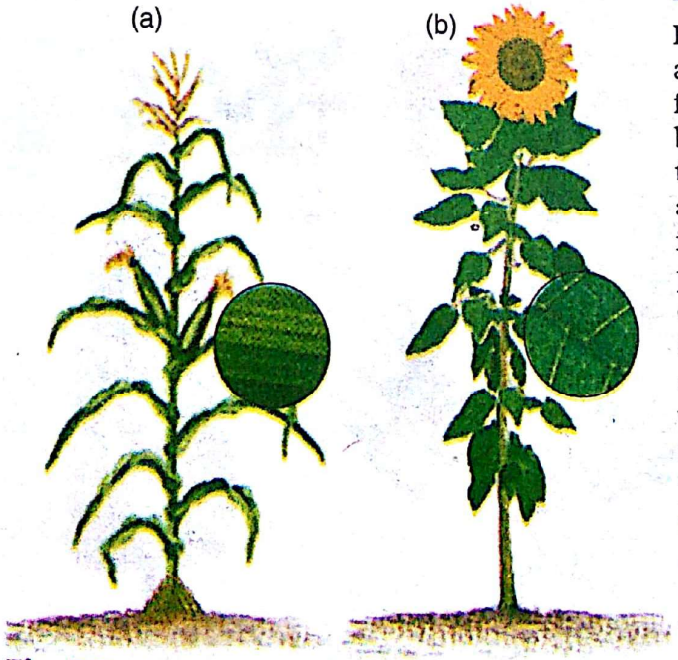

Fig. 5.6 *Angiosperms (a) Monocotyledon (Maize) (b) Dicotyledon (Sunflower)*

Classification of Angiosperms

After Linnaeus' work on classification of plants, the taxonomists realised the necessity of information on natural history of vegetation and the affinities at various levels of hierarchy. The studies were aimed at evolving natural systems of classification of plants. One of the most well-known natural systems of classification of angiosperms was proposed by two British botanists, George Bentham (1800-1884) and Joseph Dalton Hooker (1817-1911). They recorded precise description of most of the plants known at that time and classified them according to their system of classification, which also included gymnosperms. This monumental work which took about quarter of a century for compilation, was published in three volumes of *Genera Plantarum (1862 - 1883).*

Bentham and Hooker's System of Classification

Bentham and Hooker's system is based on form as well as the relationships of plants. It is still followed and used in a number of herbaria and botanical gardens all over the world. In India too, the angiospermous plants are arranged according to this system. The system being handy, it is preferred and used by the students in practical classes. Further, the generic descriptions are complete, accurate and are based on direct observations. The large genera are divided into sections and sub-sections. The various taxonomic categories discussed in Chapter 4 were assigned to all the plants published in their work. However, they could not place satisfactorily a few orders in their scheme of arrangement. These were named as **Ordines Anomali** due to their unusual position. This system covered about ninety-seven thousand species of seed plants. The phanerogams, based on their morphological characters, such as leaf

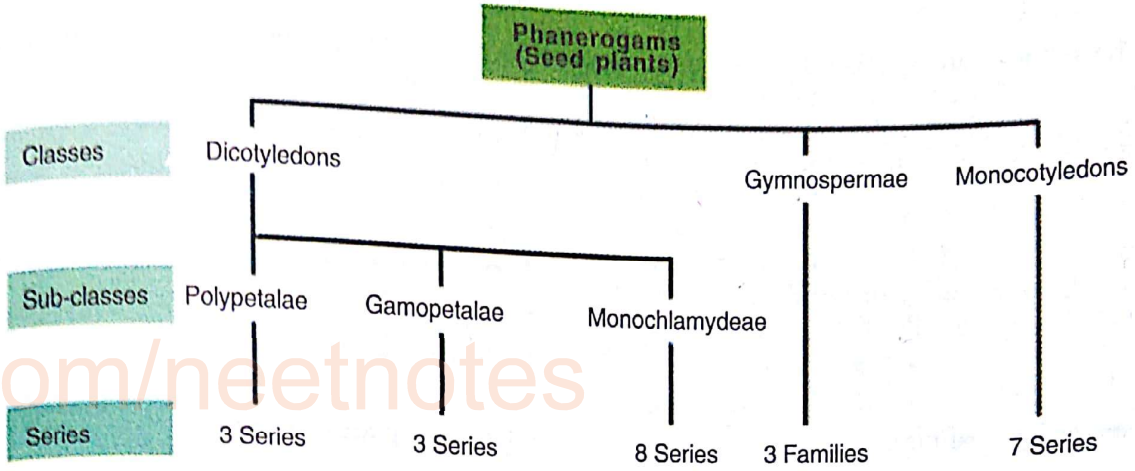

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Fig. 5.7 *Bentham and Hooker's system of classification of seed plants*

arrangement and venation pattern; number of members in floral whorls, like calyx, corolla, androecium and gynoecium; number of cotyledons in the seed and seeds with or without cover, were divided into three classes: (i) Dicotyledons (ii) Gymnospermae, and (iii) Monocotyledons (Fig.5.7).

Class I : Dicotyledons

The leaves in members of this class exhibit reticulate (net-like) venation and show varied arrangement like alternate, spiral or whorled. The flowers are tetramerous or pentamerous having four or five members in the various floral whorls, respectively. The vascular bundles are open, i.e. cambium is present between the xylem and phloem. The seeds of dicotyledons are with two cotyledons as the name indicates. Bentham and Hooker further divided them into three sub-classes, namely Polypetalae, Gamopetalae and Monochlamydeae on the basis of the number and nature of floral whorls. The three sub-classes have been further divided into series considering mainly the position of ovary with respect to the other floral parts. Series have been further categorised into 'cohorts' (equivalent to orders in other systems) and 'orders' (equivalent to families). Let us briefly study the main characters used by them to distinguish the sub-classes and series of dicotyledons.

(a) Sub-class **Polypetalae**
(*Poly* : free, *petalae* : petals)

The sepals and petals in the flowers are distinct and form two whorls. The corolla in this sub-class forms a whorl of 4-5 petals which are free from one another.

Series (i) Thalamiflorae

The sepals in this series are free from the ovary. Sepals, petals and stamens are many and arise from the thalamus below the ovary, and the flowers thus show hypogynous condition with superior ovary, no disc present below ovary. It includes orders such as, Ranales, Parietales and Malvales,

Series (ii) Disciflorae

The sepals are either free (polysepalous) or fused (gamosepalous). Flowers are hypogynous with superior ovary. A nectariferous disc surrounds the base of ovary. It includes orders *viz.*, Geraniales, Olacales, Celastrales and Sapindales.

Series (iii) Calyciflorae

The sepals are usually fused to form a tube surrounding the ovary. Flowers are perigynous or epigynous and the ovary is mostly inferior. It includes orders such as, Rosales, Myrtales and Umbellales,

(b) Sub-class **Gamopetalae**
(*Gamo* : fused, *petalae* : petals)

The sepals and petals are distinct. Again the corolla comprises 4-5 petals which are either partially or completely fused to one another. Stamens are usually epipetalous (attached to the petals). This sub-class also has three series.

Series (i) Inferae

The stamens are equal to the number of lobes of corolla. Flower shows epigynous condition and ovary is inferior. The orders are Rubiales, Asterales and Campanulales.

Series (ii) Heteromerae

Stamens are either equal or twice the number of corolla lobes and free from corolla. Ovary is superior with more than two carpels. Flower is hypogynous. The orders are Ericales, Primulales and Ebenales.

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Series (iii) Bicarpellatae

Stamens are sometimes less in number than corolla lobes, carpels usually two or more with superior ovary, indicating hypogynous condition. It includes orders Gentianales, Polemoniales, Personales and Lamiales.

(c) Sub-class **Monochlamydeae**

(*Mono* : one, *chlamydeae* : seriate or whorl)

Flowers are incomplete in this sub-class. Sepals and petals are not distinct. Flowers usually possess only one whorl of perianth which is sepaloid (petals absent). No cohorts (i.e., orders) were designated for groups of families, and in lieu of this one or more representative families are cited as examples. The sub-class has eight series as given below:

Series (i) Curvembryeae

Embryo curved around the endosperm, ovule usually one (Chenopodiaceae, Polygonaceae and Amaranthaceae, etc.)

Series (ii) Multiovulatae aquaticae

Ovules many, sub-merged aquatic plants (Podostemonaceae; also written as Podostemaceae).

Series (iii) Multiovulatae terrestres

Terrestrial plants with several ovules (Nepenthaceae, Aristolochiaceae,etc.).

Series (iv) Microembryeae

Embryo minute, endosperm fleshy (Piperaceae and Myristicaceae).

Series (v) Daphnales

Ovary usually with one carpel and a single ovule (Laurineae, Proteaceae, etc.).

Series (vi) Achlamydosporeae

Ovary usually inferior, unilocular with 1-3 ovules (Loranthaceae, Santalaceae, etc.).

Series (vii) Unisexuales

Flowers unisexual (Euphorbiaceae, Platanaceae etc.).

Series (viii) Ordines anomali

Families of uncertain relationships (Ceratophyllaceae, Salicaceae and Empetraceae, etc.).

Class II: Gymnospermae

This class has been placed between the dicotyledons and monocotyledons. The members of this class have naked ovules or seeds. Some other characters of gymnosperms have been mentioned earlier. It includes three families – Cycadaceae, Coniferae and Gnetaceae.

Class III: Monocotyledons

(*Mono* : one, *cotyledons* : cotyledon)

The leaves are simple with a parallel venation. The vascular bundles are closed (cambium absent) and scattered in the parenchyma. The flowers are trimerous having 3 members in each floral whorl. The seeds possess only one cotyledon. The monocotyledons have seven series, on the basis of the nature of perianth and condition of the ovary. These are:

Series (i) Microspermae

The perianth is petaloid with inferior ovary showing epigynous condition of the flower. The seeds are small and exalbuminous or non-endospermic i.e., without endosperm (Orchidaceae).

Series (ii) Epigynae

The perianth is partly petaloid with generally inferior ovary as the name suggests. Seeds are endospermic with nutritive tissue (Iridaceae).

Series (iii) Coronarieae

The flower has petaloid perianth with superior ovary. The seeds are endospermic (Liliaceae).

Series (iv) Calycineae

Perianth in the flowers is sepaloid with superior ovary and endospermic seeds (Palmae).

Table 5.1 Comparison of Dicotyledons and Monocotyledons

Character	Dicotyledons	Monocotyledons
Morphology	Tap roots Reticulate venation Tetra- or pentamerous flowers	Adventitious roots Parallel venation Trimerous flowers
Anatomy	Vascular bundles arranged in a ring numbering 2- 6 Open and with cambium	Vascular bundles scattered in the ground tissue, many in number Closed and without cambium

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Series (v) Nudiflorae

Perianth is either totally absent or scaly. Ovary superior, endospermic seeds (Typhaceae).

Series (vi) Apocarpae

Perianth shows either two whorls or sometimes it may be totally absent. Carpels free, ovary superior, seeds are non-endospermic (Alismaceae).

Series (vii) Glumaceae

Perianth is either absent or scaly and highly reduced. Ovary is unilocular and has a single ovule Grammineae (Cyperaceae, Poaceae).

The distinguishing features of the monocotyledons and dicotyledons are given in Table 5.1.

Merits of Bentham and Hooker's System of Classification

Bentham and Hooker's system of classification of flowering plants was based on the actual examination of specimens and preserved herbarium sheets. The descriptions were quite accurate not only for the individual species but also for the families and the genera described in the system. These descriptions of plant species were easy to follow and were of practical utility for identification of species up to the family level. Further, they had taken care to provide information related to geographical distribution of various genera. Although their system represents a natural and not a phylogenetic approach, some aspects of the system does show affinity with modern concepts of evolution. For example, the **Order Ranales** is placed in the beginning of the arrangement, and this has now been established amongst the most primitive orders on the basis of recent taxonomic findings. The placement of monocots after the dicots (from which they have been derived) also appears to be in accordance with the evolutionary trends.

Drawbacks

There are some drawbacks also in this system. For example, the position of Gymnospermae in between dicots and monocots has not been acceptable and satisfactory. The system also does not take into account several important floral characters and neglects any evolutionary considerations of genus, family and order. Further, some closely related families are placed apart and Monochlamydeae as a sub-class has been found to be an artificial group. Families like Chenopodiaceae and Caryophyllaceae have been paced in different classes. The placement of families like Asteraceae in the beginning of Gamopetalae and Orchidaceae in Microspermae do not justify the recent evolutionary approach. Interestingly, Darwin's study was available to Bentham and Hooker but somehow they did not consider evolutionary trends in their classification to rectify these lacunae. As such, several other systems have been proposed by the taxonomists which are based on phylogenetic relationships of plants.

SUMMARY

There are two main systems for classification of all the living organisms. The two-kingdom system includes all plants in plant kingdom whereas the animal kingdom includes all the animals. The other system is the five kingdom system. In this, all plants except some algae and fungi, have been put in the Kingdom Plantae. Plants are classified into Sub-Kingdoms-Cryptogamae and Phanerogamae. The former are flowerless whereas the latter bear flowers and seeds. The Cryptogamae is further divided into Divisions called Thallophyta, Bryophyta and Pteridophyta. The phanerogams are seed bearing plants and are divided into Gymnosperms and Angiosperms. Thallophytes comprise algae and fungi mainly distinguished on the basis of organization of the thallus, and mode of nutrition. The algae have green, red and brown forms which are autotrophs. These are further divided on the basis of nature of thallus and pigment colour. Bryophytes have a green, thalloid body. On the basis of the shape, internal structure of thallus, and reproductive organs these are divided into three classes, Hepaticopsida (Liverworts), Anthocerotopsida (Hornworts), Bryopsida (Bryopsida) and Pteridophytes have a plant body with rhizome bearing well developed roots and leaves. The vascular system is variable. The group is divided into Psilopsida, Lycopsida, Sphenopsida and Pteropsida on the basis of body organisation, nature of vascular system, and the mode of reproduction.

The seed-bearing plants were mainly grouped into gymnosperms and angiosperms. The former are with naked seeds whereas the latter bear seeds enclosed in ovaries (fruits). Both gymnosperms and angiosperms have also been put in Spermatophyta due to the presence of seeds.

Gymnosperms have well developed plant body differentiated into root, stem and leaves. The seeds are uncovered. Majority of the members have well organised cones containing Sporangia. Gymnosperms are further divided into Cycadopsida, Coniferopsida and Gnetopsida.

There are several systems of classification of the seed plants. A system based on natural form-relationships was proposed by Bentham and Hooker. This classification divided the group into 3 classes – Dicotyledons, Gymnospermae and Monocotyledons, on the basis of vegetative as well as floral features. The dicotyledons, having reticulate venation in leaves, open vascular bundles, two cotyledons in seeds, tetra-and pentamerous flowers, were further divided into 3 sub-classes - Polypetalae, Gamopetalae and Monochlamydeae, on the basis of arrangement of floral whorls in the thalamus. Each sub-class has series, orders and families. Monocotyledons generally have parallel venation in leaves, a single cotyledon in the seeds and trimerous floral whorls. The vascular bundles are closed and scattered in the ground parenchymatous tissues. Monocots have been divided into 7 series which are then divided into families.

The classification of phanerogams by Bentham and Hooker based on description of herbarium and live specimens has been found to be of great taxonomic utility. This, being a natural system, has some affinity with modern concepts of phylogenetic relationship. However, it has a serious drawback as far as placement of gymnospermae between the dicots and monocots is concerned. It is also weak on evolutionary basis of grouping of genera, families and orders, since some closely related families are placed apart.

EXERCISES

1. Distinguish between Cryptogamae and Phanerogamae.
2. List common modes of reproduction in algae.
3. Draw a sketch of a unicellular non-flagellated member of algae.
4. How algae differ from fungi?

5. Name the different pigments found in algae.
6. What is the basis of classification of algae?
7. Explain briefly the alternation of generation in bryophytes.
8. Describe the main features of pteridophytes.
9. Name the four classes of pteriodophytes.
10. Explain the nature of sporophylls in pteridophytes.
11. Describe the important characteristics of gymnosperms.
12. How would you distinguish monocots from dicots?
13. Name two characters used for the classification of dicotyledons into 3 sub-classes.
14. Which are the three Series of Sub-class Gamopetalae?
15. Monochlamydeae is also called Incompletae. Why?
16. What criteria are mainly used for dividing Monochlamydeae into Series?
17. Write merits and demerits of Bentham and Hooker's System of classification of seed plants.
18. Define the term Trachaeophyta.
19. Match the plants (column I) with the plant groups (column II)

Column I	Column II
Plants	**Groups**
(a) *Chlamydomonas*	(i) Angiosperm
(b) *Cycas*	(ii) Pteridophyte
(c) *Adiantum*	(iii) Algae
(d) *Rosa*	(iv) Gymnosperm
	(v) Fungus

20. Fill in the blanks.
    (i) The yellow or brown spots which have sporangia in ferns are called_____.
    (ii) Cones represent the _____ organs in the gymnosperms.
    (iii) Trimerous condition of floral whorls is characteristic of _____.
    (iv) The gymnosperms are _____ seeded plants whereas the angiosperms are ____.
    (v) The _____ are root-like structures which help in anchorage and absorption of water in the bryophytes.

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CHAPTER 6

CLASSIFICATION OF ANIMALS

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You have learnt that classification is the ideal means of studying the diversity of organisms. You are also familiar with the systems of classification, in general (Chapter 4). The two-kingdom classification divides the living organisms, *viz.* all plants into 'Plantae' and all animals into 'Animalia'. With the availability of more information about microorganisms, biologists have added kingdoms and grouped the unicellular plants and animals together into 'Protista'. In this system, 'Plantae' contains only the multicellular plants and 'Animalia' contains only the multicellular animals. You have learnt about various plant groups in the previous chapter. This chapter will help you to acquaint with the important characteristics relevant for classification of animals, including the unicellular protozoan protists.

6.1 SOME GENERAL FEATURES OF ANIMALS

Animals make up millions of species and are among the most beautiful living things. They are very diverse in form, ranging from single-celled microscopic ones (*Amoeba*) to multi-cellular macroscopic whales and giant squids.

Grades of Organisation and Body Plan

Animals though show different shapes and sizes but possess either cellular, tissue, organ or organ system of organisation. The cellular grade of organisation can be observed in all protozoans (unicellular or acellular) where all the vital activities of the body are performed by a single cell, for example *Amoeba*. Animal cells are diverse in structure and function, lack a rigid cell wall and are quite flexible. The next higher level of body organisation is observed in multicellular animals, which are called **metazoans**. Based on complexity of organisation, metazoans are further subdivided into two subkingdoms, the **Parazoa** and **Eumetazoa**. In Parazoa (e.g. sponges), the cells are loosely aggregated and do not form tissues or organs. In Eumetazoa, which includes the rest of the animals, the cells are organised into structural and functional units called tissues, organs and organ systems. The body plan of eumetazoans resembles either a blind sac (cnidarians like *Hydra* and platyhelminthes) or a tube within a tube plan (rest of eumetazoans). Figure 6.1 shows the different body plans.

Symmetry

The parazoan animals, sponges, lack any definite symmetry. The eumetazoan animals can be grouped into two categories based on symmetry (Fig. 6.2). When any plane passing through the central axis of the body divides the organism into halves that are approximately mirror images it is called **radial** symmetry and the animals showing this symmetry are called **Radiata**. For example, cnidarians (sea anemone, jellyfish and corals) and ctenophorans (comb jellies) have this kind of body plan. These animals have an oral and the opposite aboral ends but no left and right sides. In the remaining eumetazoans, the body can be divided into identical left and right halves in only one plane. This kind of symmetry is called bilateral symmetry and such animals are called **Bilateria**. These animals, for example fish, have a front or anterior end and a rear or posterior end, an upper back or dorsal side and a lower belly or ventral side.

Diploblastic and Triploblastic Organisation

In radiata, the cells are arranged into two fundamental layers, an external ectoderm and

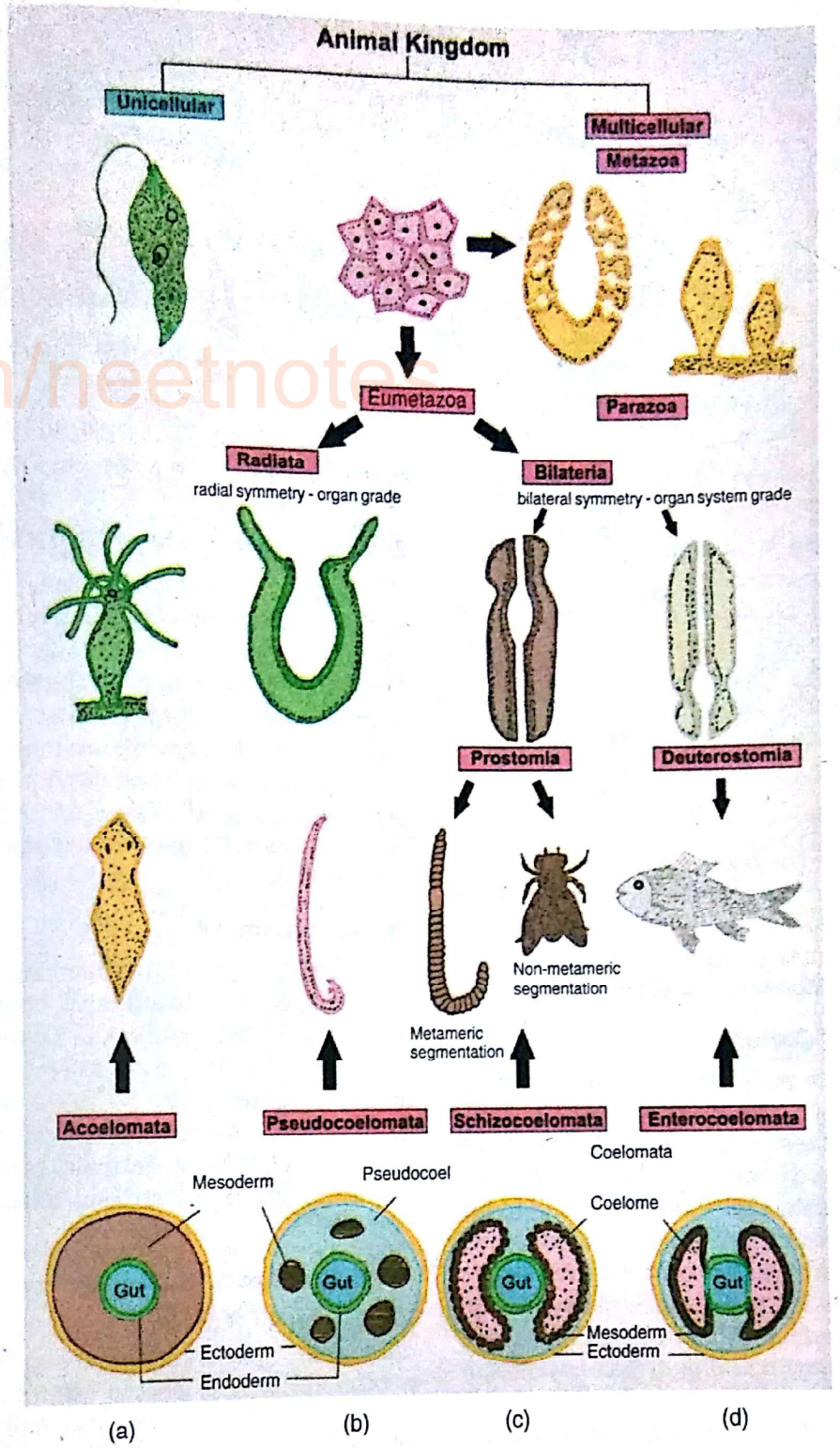

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Fig. 6.1 *Diagrammatic representation showing body organisation (grades and symmetry) and developmental plan in protozoa and metazoa. Coelom is absent in acoelomates (a) Formation of coelom in other bilateria is shown in cross-sectional view. (b-d) In pseudocoelomata, the main body cavity is a pseudocoel. (b) Coelom formation takes place by splitting of mesodermal pouches present on the lateral sides of the prospective gut, and (c) by growth of lateral mesodermal pouches from the endoderm.*

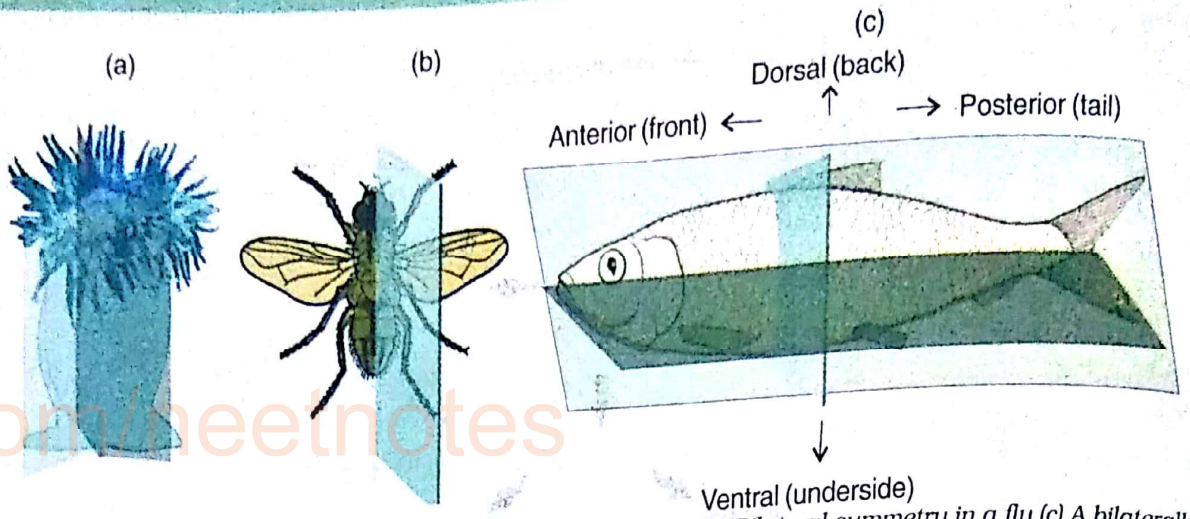

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Fig. 6.2 *Body symmetry (a) Radial symmetry in a sea anemone (b) Bilateral symmetry in a fly (c) A bilaterally symmetric (fish)*

an internal endoderm with an intervening mesoglea. Such animals are, therefore, called **diploblastic**. In Bilateria, a third germ layer, mesoderm, is present in between the ectoderm and endoderm. Hence, they are called **triploblastic** animals. In these animals, embryonic blastopore forms mouth in prostomia and anus in deuterostomia.

Segmentation

In some Bilateria the body is of many segments, which show serial repetition of parts, for example, earthworm. This kind of segmentation is called metameric segmentation and the phenomenon is known as **metamerism.**

Body Cavity or Coelom

The building plan of Bilateria is achieved from the three primary germ layers of their embryos, *viz.*, ectoderm, mesoderm and endoderm. The space between body wall and alimentary canal (body cavity) remains lined by mesoderm and is called coelom. Visceral organs lie in the coelom. The animals in which the coelom is absent are called **acoelomates**, for example flatworms. In them, the space between ectoderm and endoderm is filled with parenchyma derived from mesoderm. Still in some Bilateria, the body cavity is not lined by mesoderm. Instead, the mesoderm is present as scattered pouches in between the ectoderm and endoderm. Such a body cavity is called pseudocoelom and the animals (e.g. roundworms) possessing pseudocoelom are called **pseudocoelomates** (See Fig.6.1).

Heterotrophic Mode of Nutrition

Unlike plants, which synthesise their own food and are called **autotrophs**. Animals that feed upon other living forms are called **heterotrophs**. The heterotrophs may be **herbivores** (eating plants), **carnivores** (eating animals) and **omnivores** (eating both animals and plants). There are animals that depend on other organisms for food. These are called **parasites** and the relationship is known as host- parasitic relationship.

Active Movement

Compared to members of other kingdoms, animals perform a more rapid and complex way of movement called locomotion. Some animals can fly (butterflies, bats and birds). Some can swim (jellyfish, squids, fishes, whales) and others can run or walk on land. Movement of animals is directly related to the flexibility of their cells, and perhaps, the most striking characteristic of animals.

Reproduction and Development

Most animals reproduce sexually. Cells formed after meiosis function directly as gametes. Male produces male gametes (spermatozoa), and female produces female gametes (ova). Fusion of gametes (fertilization) results in the formation of a zygote. As a result of sequential development, zygote produces a complete organism.

6.2 CLASSIFICATION OF ANIMALS

The animal kingdom includes about 35 Phyla (*sing. Phylum*) of which 11 are considered as

major Phyla. Almost 99 per cent of animals are **invertebrates** (animals without backbone) and the remaining represents the **vertebrates** (animals with backbone). Also, the animals are categorised into two major groups, **non-chordates and chordates**, on the basis of the presence or absence of notochord at some stage in their life. We will study the important characteristics of these groups.

Phylum Protozoa
(Unicellular Protist Animals)

There are about 15,000 species of protozoans known to exist in the world. They are microscopic organisms in which a single cell performs all the vital activities. For this reason, protozoans are also referred to as acellular organisms. They are aquatic (fresh water and marine), and cosmopolitan in distribution. Some forms are parasitic. The protozoan cell body is either naked, for example, *Amoeba*, or surrounded by a non-rigid pellicle. Cellulose is absent in pellicle. Some protozoans secrete shells of various inorganic compounds as external covers (foraminiferans). Different types of locomotory organs are found in protozoans. They may bear flagella (flagellates), or cilia (ciliates) (Fig.6.3), pseudopodia (sarcodines).

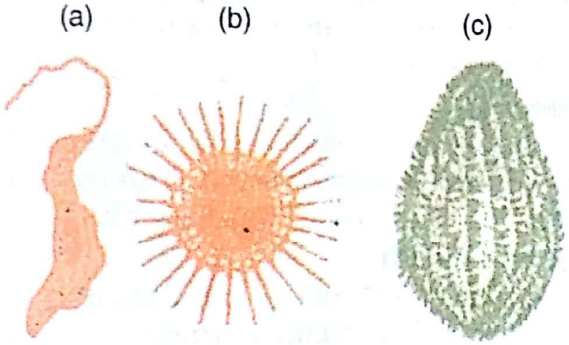

Fig. 6.3 *Some examples of protozoans with different types of locomotory organs: (a) Trypanosoma having flagellum (flagellate) (b) Helizoan bearing cilia (ciliate) (c) Tetrahymena having cilia*

Locomotory organs are absent in the parasitic forms (Sporozoa). In them, neurofibrils and contractile myofibrils are present underneath the cell surface. Most protozoans are free-living and aquatic. They are holozoic and feed largely on bacteria, microscopic algae and minute animals such as rotifers or on other protozoans including members of their own species. Some protozoans are holophytic; they contain chlorophyll and prepare their own food by photosynthesis (e.g. *Euglena*). The parasitic protozoans feed on materials obtained from the hosts (e.g. *Monocystis*).

Contractile vacuole is found in almost all fresh-water protozoans for maintenance of osmotic concentration of cell body, the phenomenon is known as **osmoregulation**. Contractile vacuole also helps in excretion. Many sporozoan parasites are relatively harmless, but some are harmful also. For instance, *Plasmodium vivax* and *Plasmodium falciparum* cause malaria in humans. Protozoans are generally uninucleate, but ciliates and many amoeboid types are multinucleate.

The pattern of reproduction is also very specialised in different protozoans. Most sarcodines, flagellates and ciliates show asexual reproduction by binary fission, multiple fission or even budding. Some ciliates, for example, *Paramoecium* reproduce by sexual means in which two individuals come close to each other and interchange genetic material by a process known as **conjugation**. There is no gamete formation in such a process. In sporozoa, some stages of life cycle show formation of gametes, which are morphologically distinct.

Examples

Free living - *Euglena*, *Amoeba*, *Paramoecium*, *Noctiluca* and *Elphidium*.

Parasitic - *Monocystis*, *Entamoeba*, *Plasmodium*, *Trypanosoma* and *Giardia*.

Phylum-Porifera
(Pore Bearing Animals)

Members of this Phylum are commonly known as sponges. They are the most primitive group of multicellular animals. About 5000 species of sponges are known. Most of them are marine and remain attached to rocks (sessile). A few live in fresh water. Some sponges have been found in rocks which are more than 600 million years old. They range from 1 cm to 1 metre in length. Some are radially symmetrical, but the larger ones are asymmetrical. They are multicellular and represent cell aggregate body plans. As they have no tissue grade of organisation they are included in the subkingdom Parazoa (Fig. 6.4).

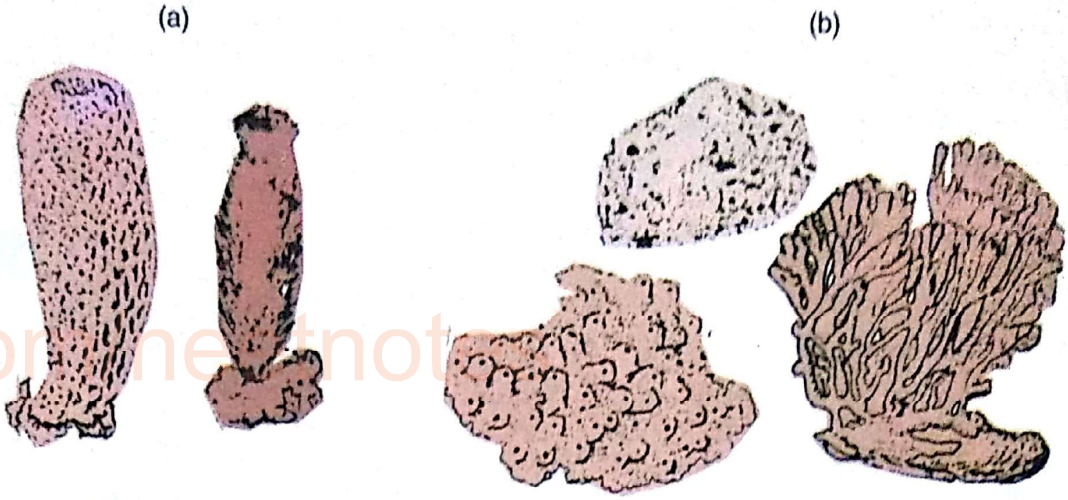

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Fig. 6.4 *Shapes of some sponges : (a) radially symmetric and (b) asymmetric forms*

Sponges are sessile, grow into various shapes (Fig. 6.4), and are attached to an under-water object, except the fresh water sponges. Numerous pores connect the outside to a central chamber (Fig. 6.5). Several minute pores (ostia; sing. ostium) present on the body lead into canals (canal system) lined by flagellated collar cells (choanocytes). At the terminal point of the body a large aperture called osculum is present. Water along with food enters the canals through the ostia and comes out along with excretory materials, sperm and ova through the osculum. Sponges are diploblastic. They also contain amoebocytes, pinacocytes and other cell types.

The body is supported by an internal skeleton formed of innumerable calcareous or siliceous spicules or proteinaceous spongin fibres.

Sponges reproduce asexually by fragmentation. They exhibit great power of regeneration. During sexual reproduction some cells become egg or sperm cells. After fertilisation, the zygote develops into a flagellated larva, which swims, settles in a new place and grows into a sponge.

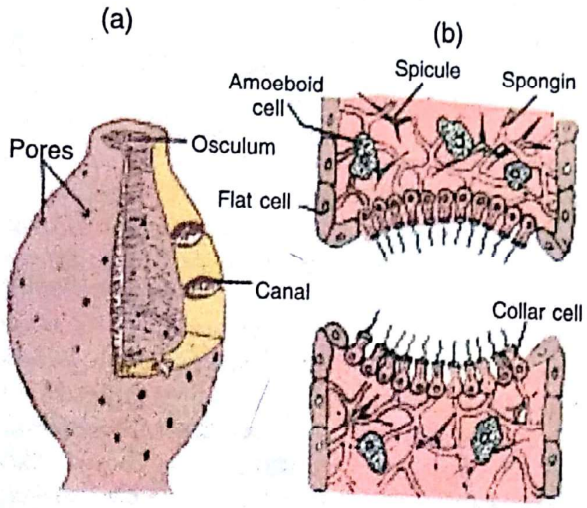

Fig. 6.5 *(a) A cut-away diagram of a simple sponge showing pores, canal, osculum and internal central chamber (b) Cross-section of a sponge wall showing flagellated collar cells lining a pore, flat and amoeboid cells and matrix material*

Examples

Sycon (Scypha), *Spongilla*, *Potorion* (Neptune's cup), *Chalina* (Dead man's finger), *Euspongia* (Bath sponge) and *Euplectella* (Venus flower basket).

Phylum Cnidaria

The phylum Cnidaria includes about 9,000 species, which are mainly marine; few are fresh water. The phylum name is derived from the stinging cells or cnidoblasts present on the ectoderm of tentacles and body of these carnivorous animals. Cnidarians have achieved tissue grade of organisation and they exhibit a blind sac body plan and radial symmetry (Fig. 6.6).

Cnidarians are diploblastic animals in which the body wall consists of only two layers of cells, an outer ectoderm and an inner endoderm, separated by a gelatinous layer of mesoglea (Fig.6.7). Body contains the oral aperture, for

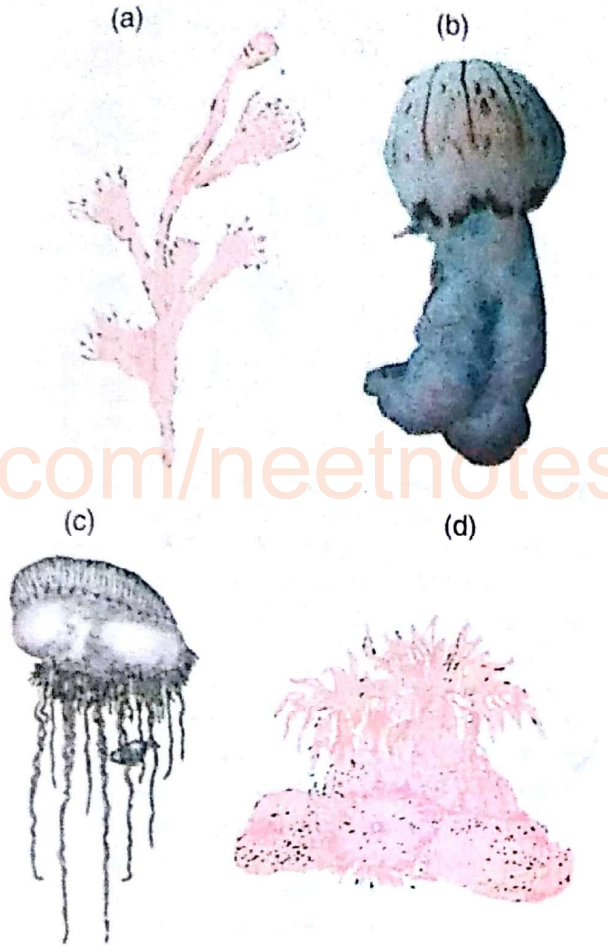

Fig. 6.6 *Some cnidarians (a) Obelia (b) Jellyfish (c) Physalia (d) Sea anemone*

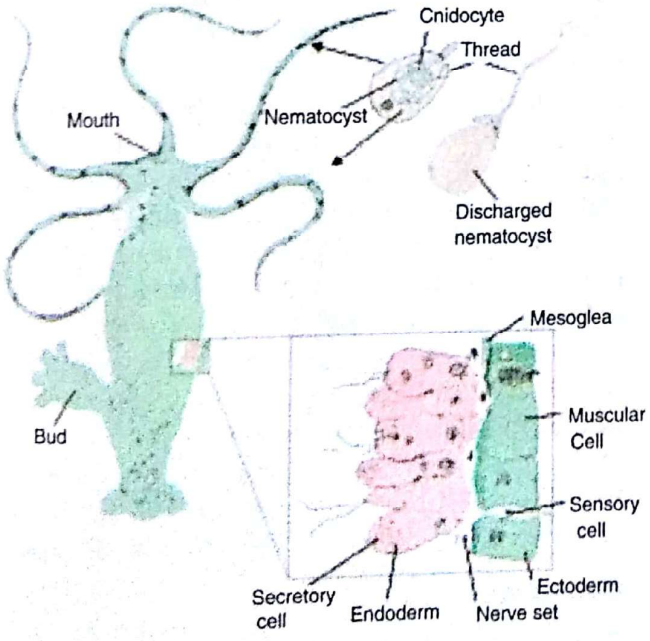

Fig. 6.7 *Structure of Hydra*

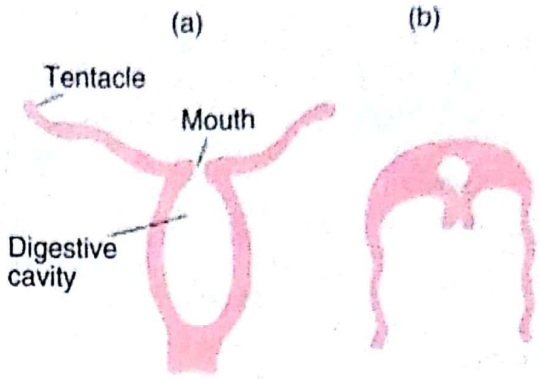

Fig. 6.8 *Polyp and medusa body forms (a) sessile polyp (b) swimming medusa*

ingestion and egestion, leading into a gastrovascular cavity and anus is absent. Such an enteron with single opening is called coelenteron hence the previous name of the phylum was Coelenterata which used to include the Cnidarians along with Ctenophores. At present, the Ctenophores have been assigned a separate phylum status.

The endodermal cells lining the gastrovascular cavity are specialised for secreting digestive enzymes. These enzymes partly digest food by extracellular digestion, which is then absorbed as solution and ingested as small particles as well, by the endoderm. Undigested material is egested through the mouth.

Cnidarians exhibit two basic body forms, the **polyp** and **medusa** (Fig. 6.8). The polyp is sessile, solitary or colonial and resembles a cylindrical stalk with mouth and tentacles facing upwards. The medusa is a solitary and free-swimming. It is like a bell or an umbrella with mouth and tentacles facing downwards. The medusa can be regarded as an upside-down polyp with reduced stalk, which can swim away. In many cnidarians, polyps give rise to medusae by vegetative budding, and medusae form polyps following sexual reproduction. Cnidarians like *Obelia*, pass through both polyp and medusa stages in their life cycles (dimorphism). Polyps reproduce asexually by budding whereas medusae liberate gametes into water during sexual reproduction. Both asexual and sexual forms are diploid and the only haploid cells are gametes. Such alternation of asexual and sexual phases in the life cycle of Obelia is called **metagenesis**. It should not be confused with alternation of generations as found in plants.

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Some forms show division of labour by forming structurally and functionally different types of individual (zooids) within the same organism during its life history. This is called **polymorphism**. Following fertilisation, the zygote forms a ciliated larva called planula, which swims, settles and grows into a sessile polyp. Some cnidarians, like Hydra, do not have a medusa stage.

Examples

Hydra, *Porpita*, *Vellela*, *Physalia* (Portuguese man-of-war), *Aurelia* (Jellyfish), *Adamsia* (Sea anemone), *Pennatula* (Sea-pen) and *Gorgonia* (Sea-fan).

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Phylum Ctenophora

Ctenophores are marine animals with transparent and flat or oval body shape. Polyp phase is absent in their life cycle. These are bilaterally symmetrical and devoid of cnidoblast cells. When the tentacles are present they are two in number and contain colloblast cells. These animals move by cilia, which join together to form comb plates; there are eight median comb plates. Gastrovascular cavity is branched and open to the exterior by stomodaeum. They are diploblastic animals but the mesoglea is different from that of cnidaria; it contains amoebocytes and smooth muscle cells and is comparable to a loose layer of cells. From this viewpoint, ctenophores may be considered as triploblastic. The presence of a special sense organ at the opposite end of the mouth (aboral end) is the characteristic of the members of this phylum. They reproduce only by sexual means and do not exhibit larval phase in their life cycle.

Examples

Hormiphora, *Ctenoplana* and *Beroe*.

Phylum Platyhelminthes *(Flatworms)*

There are about 13,000 species belonging to the phylum Platyhelminthes. These are dorso-ventrally flattened and, hence, commonly known as flatworms. These are mostly parasites that live in other animals, including humans. Some are free-living forms, mainly aquatic-marine or freshwater (Fig. 6.9).

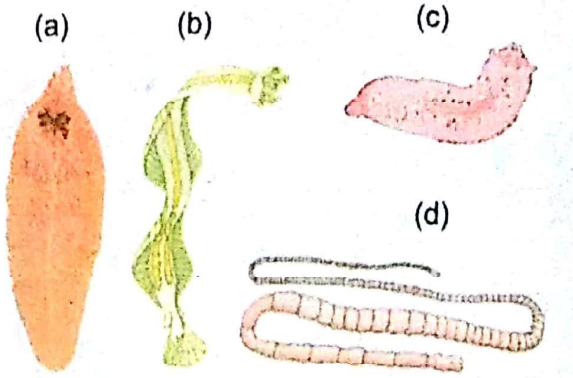

Fig. 6.9 *Some flatworms (a) Fluke (b) Turbellaria (c) Planaria (d) Tapeworm*

These are triploblastic and unsegmented animals exhibiting bilateral symmetry. They are acoelomate and parenchyma cells originating from mesoderm fill up the cavities of the body. Their bodies remain externally covered by cilia or cuticle. Platyhelminthes show organ-system grade of body organisation. Alimentary canal is incomplete having mouth opening but no anus. The parasitic forms lack alimentary canal. They absorb nutrients of the host directly through their body surface. The nervous system contains a concentration of nervous tissue in the head called the brain ganglion. A pair of ventral nerve cord leads from this ganglion posteriorly. A transverse nerve connecting the cords gives the system a ladder like appearance. These worms have an exterior cuticle instead of an epidermis. Head bears hooks and suckers. Life history usually involves intermediate hosts. Flatworms have specialised cells called flame cells, for excretion and osmoregulation.

They reproduce both asexually and sexually. Flatworms can regenerate the entire body from a body part similar to the sponges and cnidarians. The phenomenon of regeneration is asexual reproduction. They are hermaphrodite or bisexual, that is, both male and female sex cells are produced by the same individual. But the structure of body facilitates cross-fertilisation rather than self-fertilisation.

Examples

Taenia (Tapeworm), *Fasciola* (Liver fluke), *Echinococcus*, *Schistosoma* and *Planaria*.

Phylum Nemathelminthes (Aschelminthes) (Roundworms)

Phylum Nemathelminthes is represented by about 15,000 species of roundworms. These are also known as nematodes. Their bodies appear circular in cross-section, hence, the name roundworm. Though not apparent, they are possibly the most abundant among animals. Vast quantities of microscopic free-living nematodes, such as *Rhabditis*, live in soil rich in organic matter. Others are aquatic or parasitic (Fig. 6.10).

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Fig. 6.10 *Some roundworms*

Roundworms are bilaterally symmetrical, triploblastic and pseudocoelomate animals with an organ system grade of organization and tube-within-a-tube body plan. They possess elongated body with pointed ends. The digestive tract is differentiated into mouth, pharynx, intestine and anus (Fig.6.11).

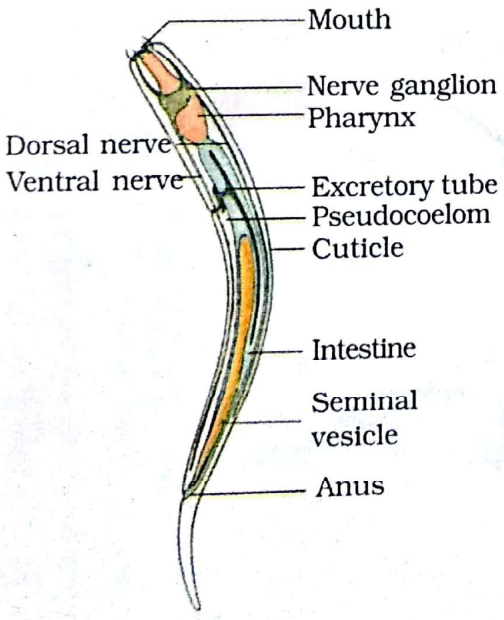

Fig. 6.11 *Structure of a roundworm (male)*

The mouth may be armed with teeth to cut and pierce tissues. The muscular pharynx allows the parasitic nematode to suck blood from the host. The body wall has longitudinal muscles and an elastic cuticle. The false body cavity allows body wall muscles and digestive tract muscles to act independently of each other. Roundworms have a flexible body movement. An organised nerve ganglion is present around the pharynx with dorsal and ventral nerves to co-ordinate movement. An excretory tube removes body wastes from the body cavity. Sexes are usually separate (dioecious) and morphologically distinct; often females are longer than the males. Fertilized eggs have a thick wall and can survive in adverse conditions.

Examples

Ascaris, *Wuchereria* (Filaria worm), *Ancylostoma* (Hookworm), *Enterobius* (Pin worm) and *Rhabditis*.

Phylum Annelida

The phylum Annelida includes over 9,000 species of metamerically segmented animals with a true coelom. The coelom too is partitioned segmentally by peritoneal membranous septa. Annelids are triploblastic and bilaterally symmetrical animals with organ-system grade of organisation. Each segment is externally divided into many parts by rings (Latin, *annulus*) and, hence, the name Annelida (Fig. 6.12).

The body is covered by definite cuticle secreted from the ectoderm. The body wall of annelids has both longitudinal and circular muscles. These contract alternately against the fluid-filled septa during locomotion. Bristles or setae on the lower side help to grip the ground during locomotion. Polychaetes (*Neries*) have numerous setae on lateral appendages called parapodia. Leeches being ectoparasites do not possess either setae or parapodia and swim by undulatory movement by musculature. The digestive system of annelids is more advanced than nematodes and contains distinct mouth and anus at opposite ends of the body. It consists of a muscular pharynx to swallow food, an oesophagus to carry it to the stomach where it is churned and digested, and a long intestine to absorb nutrients. Undigested waste is

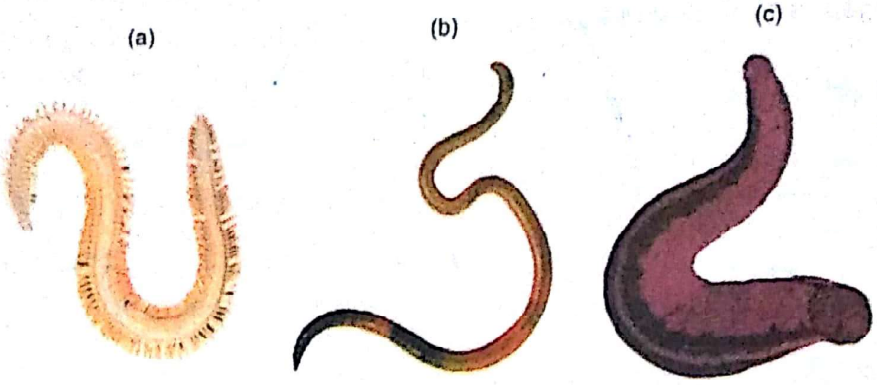

Fig. 6.12 *Some annelids: (a) Nereis (b) Earthworm (c) Leech*

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compacted and expelled as worm castings through the anus. A closed circulatory system of blood vessels, a heart to pump the blood is found for the first time in annelids. The main blood vessels, one dorsal and one ventral, have branches in each body segment which supply blood to the skin, muscles, intestines and other organs. Oxygenation of blood occurs through the moist skin. Therefore, the earthworm can live only in a moist condition. The excretory organs are paired nephridia (sing. nephridium) in each segment. They are special tubes, which collect excretory waste from the coelom to the outside. They also help in maintaining water balance of the body (osmoregulation). Body action is co-ordinated by a pair of cerebral ganglia (sing. ganglion), which are collection of nerve cell bodies. These are present at the anterior end above the pharynx and connected by lateral nerves to a double ventral nerve cord. Sexes are separate in polychaetes but earthworms and leeches are hermaphrodites.

Examples

Nereis, *Aphrodite* (sea mouse), *Pheretima* (Earthworm), *Tubifex*, *Hirudinaria* (Leech), *Chaetopterus*, *Terebella*, and *Bonnellia*.

Phylum Mollusca (Soft Bodied Animals)

Phylum Mollusca (L. *Mollis* : soft-bodied) is the second largest animal phylum which includes over 60,000 species. Molluscs form an ancient group, they have lived over 500 million years. They are triploblastic coelomates and usually with bilateral symmetry. They are terrestrial, marine or fresh water inhabitants (Fig. 6.13).

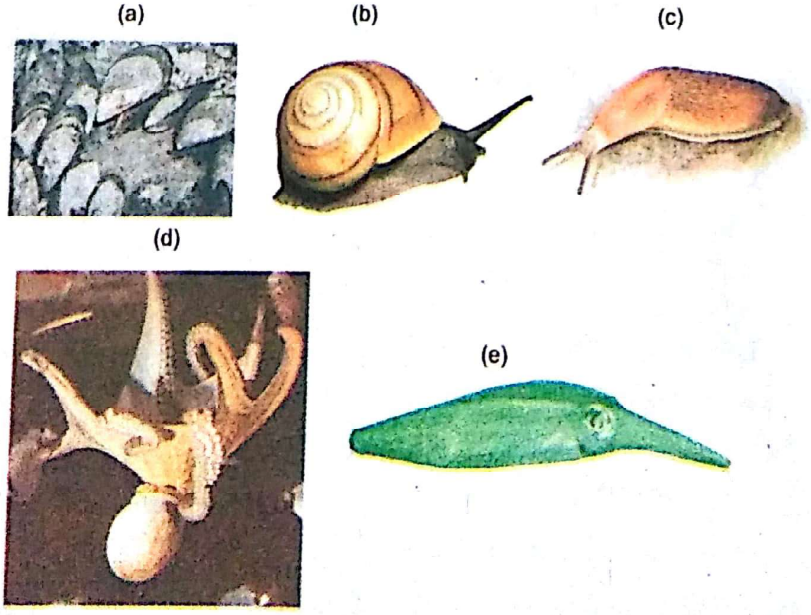

Fig. 6.13 *Some molluscs: (a) Unio (b) Snail (c) Shell-less slug (d) Octopus (e) Squid*

The body of molluscs is soft but protected by hard calcareous shell.

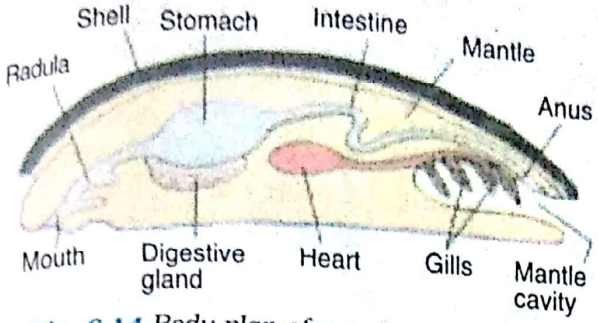

Fig. 6.14 Body plan of a mollusc (hypothetical)

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Figure 6.14 shows body organisation of a typical mollusc. The body is unsegmented and differentiated into a head, a ventral soft and muscular foot, and a dorsal visceral hump. The skin is soft and it forms a mantle over the hump. The visceral hump contains the main digestive and circulatory organs. Below the mantle, it has a number of feather-like gills. Gills have respiratory and excretory functions. The anterior head region has sensory tentacles. The mouth contains a file-like rasping organ for feeding, called radula. Molluscs are basically oviparous and development is through a trochophore larva.

Examples

Pila (apple snail), *Achatina* (land snail), *Lamellidens* (mussel), *Pinctada* (pearl oyster), *Sepia* (cuttlefish), *Loligo* (squid), *Octopus* (devilfish), Doris (sea-lemon), *Aplysia* (sea-hare) and *Teredo* (shipworm).

Phylum Arthropoda
(Animals with jointed appendages)

The phylum Arthropoda (L. *Arthros* : jointed; *podos* : legs or appendages) constitutes the largest group of animals with about 900,000 species. These are triploblastic, coelomate and bilaterally symmetrical animals (Figs. 6.15, 6.16). The body of arthropods is covered by chitinous cuticle, which forms the exoskeleton. Arthropods have a segmented body, each segment bearing a pair of jointed appendages covered by a jointed exoskeleton. The segments are not separated by septa as in annelids. The body consists of head, thorax and abdomen, in some cases, head and thorax may be fused to form cephalothorax. The head consists of several fused segments with appendages modified to serve as antennae (feelers), mouthparts or pincers (chelicerae). Arachnids have no

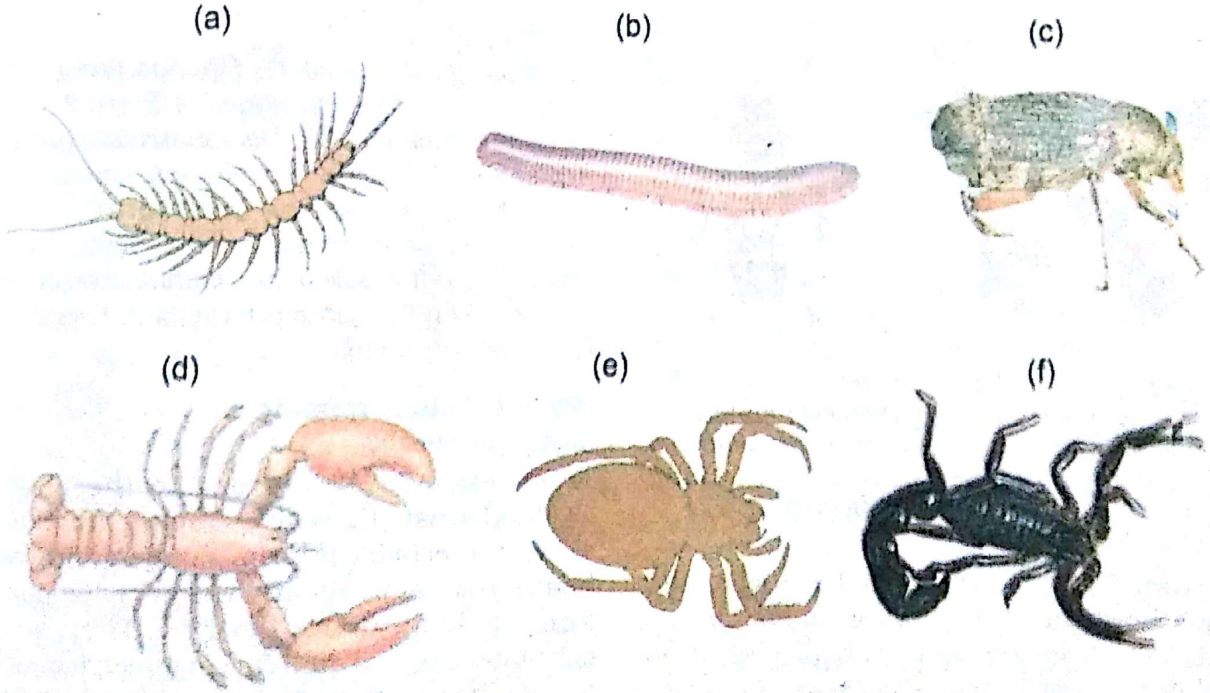

Fig. 6.15 Some common arthropods : (a) Centipede (b) Millipede (c) Beetle (d) Prawn (e) Spider and (f) Scorpion

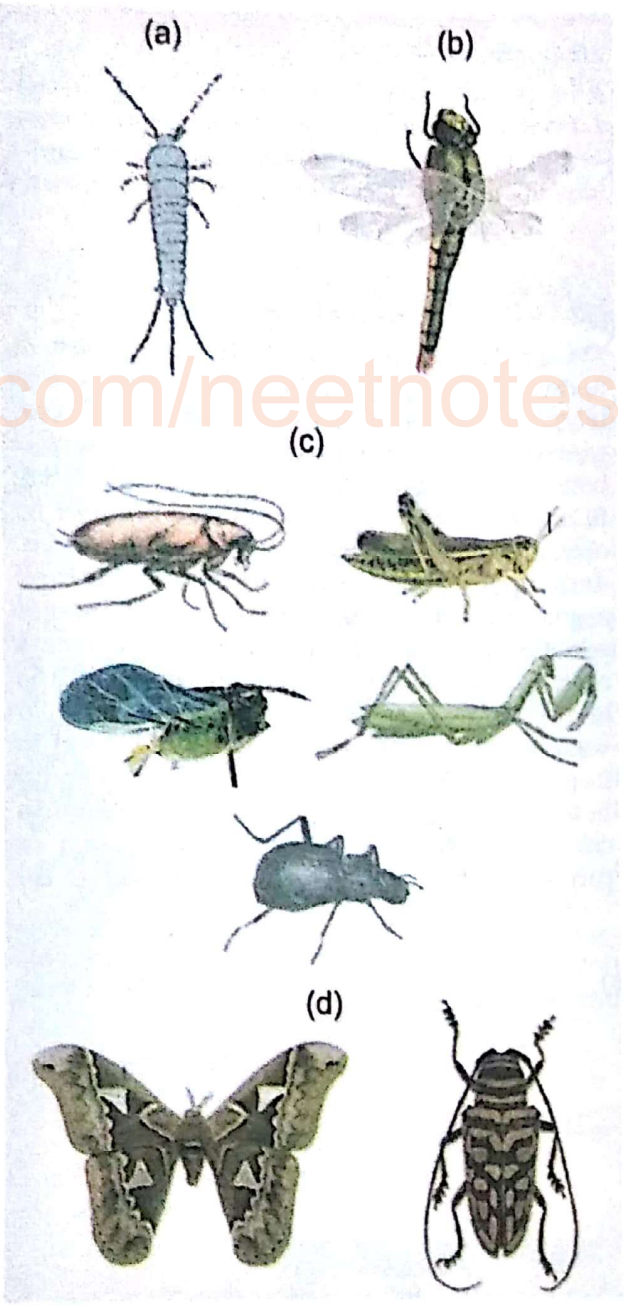

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Fig. 6.16 A few common insects (a) Silverfish (wingless form) (b) Dragonfly (wings non-foldable) (c) Cockroach, grasshopper, praying mantis, aphid (wings foldable) Bedbug (wingless) (d) Butterfly, beetle

antennae. Thoracic segments have legs (and wings); the abdomen has no legs in insects.

Respiratory organs are gills, book gills, book lungs, or tracheal system. Excretion takes place through green glands or malpighian tubules since nephridia are absent. Sensory structures in arthropods are antennae for perceiving odour, eyes, statocysts or balance organs, receptors for taste (located in their feet in insects) and sound receptors (in chirping crickets and cicadas). The arthropod eyes may be simple or compound. The heart is dorsal and circulatory system is open. The central nervous system consists of paired preoral ganglia connected by commissures to a solid ventral nerve cord, which is double and contains segmental ganglia and nerves.

Arthropods are unisexual. In a few aquatic arthropods, fertilisation is external, that is male and female sex cells fuse outside the body in water. But in others fertilisation is internal; the male deposits the sperm within the female sex organ. In all land arthropods, fertilization is always internal. Eggs are laid by most arthropods. They are oviparous. In some, like the scorpion, the eggs hatch within the female body. They bring forth the young alive. They are viviparous. In many arthropods, development is direct. The young, hatched from eggs, resemble the adult and often occupy the same habitat. They grow by moulting. In others, the egg hatches into an independent larva, which does not resemble the adult; there are many larval forms. The process of transformation of a larva into an adult is called **metamorphosis**. Larvae and adults may show a different feeding habit and occupy different habitats.

Examples

Araneus (garden spider), *Limulus* (king crab), *Buthus* (scorpion), *Eupagurus* (hermit crab), *Cancer* (common crab), *Macrobrachium* (prawn), *Lepisma* (silverfish), *Periplaneta* (cockroach), *Apis* (bee), *Anopheles* (mosquito), *Musca* (housefly), *Leptocorisa* (paddy pest: gandhi poka) *Triops* (tadpole fish), *Daphnia* (water flea), *Cyclops*, *Squilla*, *Astacus* (crayfish), *Lepas*, *and Balanus* (barnacle).

Phylum Echinodermata
(Spiny Animals)

About 6000 species belong to the phylum Echinodermata (*L. echinos* : spiny, *dermatos* : skin); the ectoderm bears many spines and, hence, the name. All are marine, triploblastic and coelomate (Fig. 6.17). The adult echinoderms are radially symmetrical with body parts occurring in five axes (pentamerous radial symmetry). Larvae are bilaterally symmetrical. The exoskeleton is calcareous consisting of many plate-like structures called

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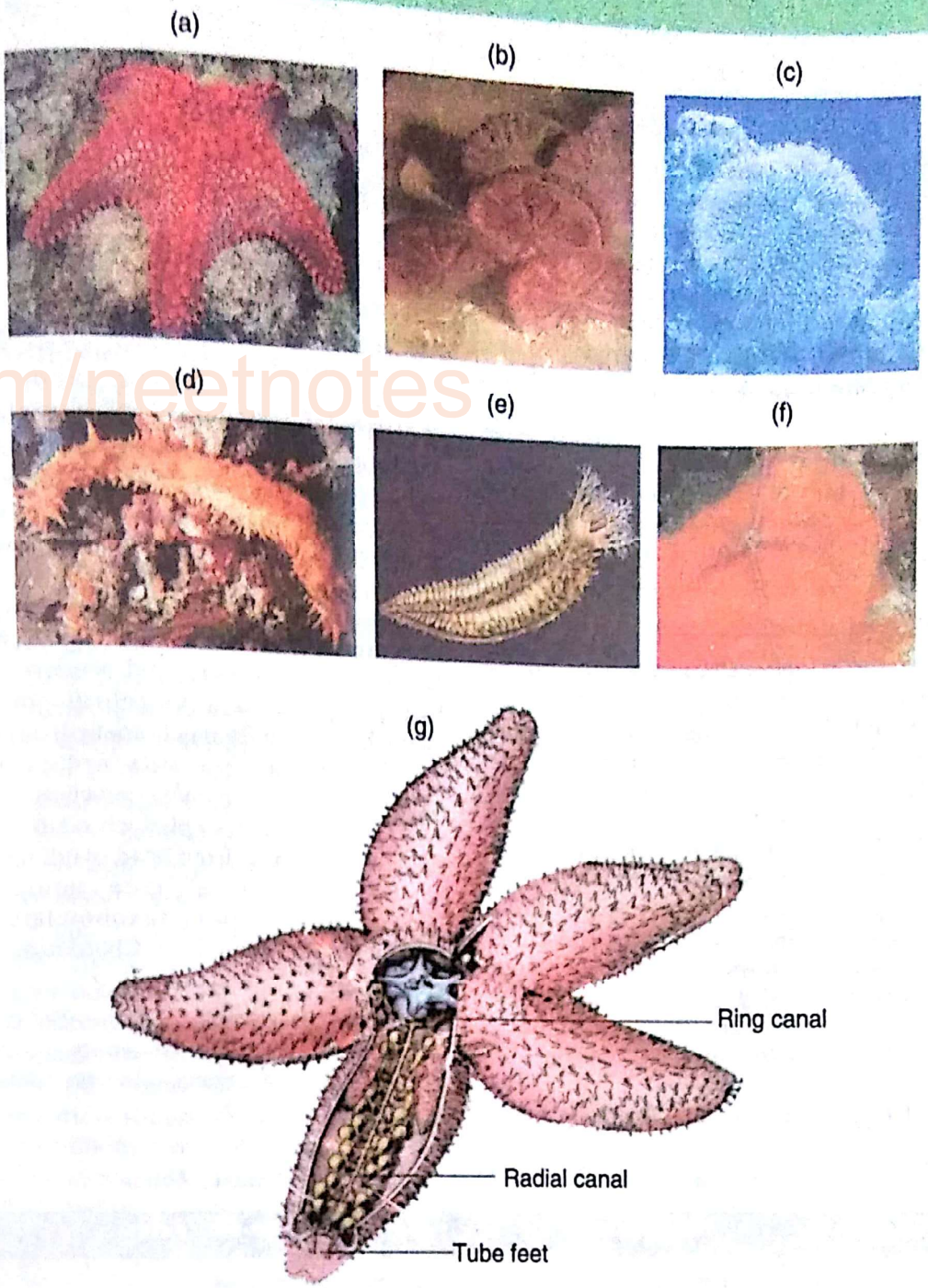

Fig. 6.17 Some echinoderms (a) Starfish (b) Sand dollar (c) Sea urchin (d) Holothuroid (e) Sea cucumber (f) Brittle star (g) Cut-away diagram to show the water-vascular system of a star fish

ossicles. They have a mouth on the lower side and anus on the upper side. The most distinctive feature of echinoderms is the presence of water vascular system, which is a part of the coelom. It consists of an array of radiating canals and tube-like appendages called tube feet (Fig. 6.17g). Their main function is locomotion and the capture of food. The tube feet also serve as the equivalent of gills during respiration. These animals have no proper circulatory system. Encircling the oesophagus and situated close to the ring canal is a nerve

ring from which emanate five radial nerves, one to each arm. Excretion is achieved partly by diffusion through the body surface, partly by amoeboid cells in the coelomic fluid. These cells absorb excretory waste and carry it to the exterior through the skin and gills. Sexes are separate with five pairs of sex organs, one pair in each arm. In most cases fertilisation occurs in open water. The development includes a free swimming larva. This larva undergoes a very complex metamorphosis into a young radial adult.

Examples

Asterias (star fish or sea star), *Echinus* (sea urchin), *Echinocardium* (heart urchin), *Antedon* (feather star or sea lily), *Cucumaria* (sea cucumber)*and Ophiura* (brittle star).

Phylum Chordata (Chordates)

The taxonomic category **Chordata** refers to the group of animals, which possess notochord either throughout or during early embryonic life. Thus, the animals without notochord are referred to as nonchordates. Notochord is a stiff and flexible rod of tissues lying ventral to nerve cord. All the chordates are triploblastic, coelomate and bilaterally symmetrical. They possess a post anal tail and closed blood vascular system. The other common features of chordates are: a dorsal hollow nerve cord and paired pharyngeal gill slits. These chordate characters are shown in the diagrammatic representation of a hypothetical chordate in Fig. 6.18.

Table 6.1 presents a comparison of salient

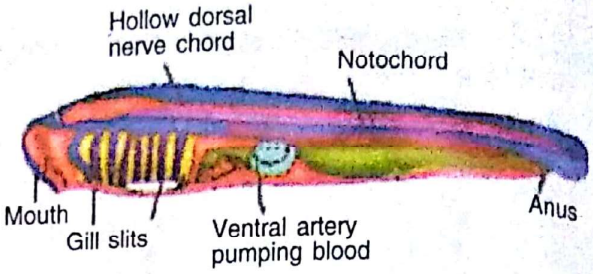

Fig. 6.18 *Internal organization of a hypothetical chordate.*

features of chordates and non-chordates. Phylum Chordata is divided into four subphyla, subphylum **Hemichordata** or **Stomochordata**, subphylum **Urochordata** or **Tunicata**, subphylum **Cephalochordata** or **Acrania** and subphylum **Vertebrata**. The first three subphyla are considered primitive and often referred to as **protochordates** or **nonvertebrate chordates**. All of them are marine and possess notochord but never form a vertebral column. In Hemichordata or Stomochordata, true notochord is absent but gill slits are present; in Urochordata or Tunicata, notochord is present only in larval tail; in Cephalochordata or Acrania notochord extends from head to tail region. Due to the absence of true notochord in Hemichordata many taxonomists do not consider these animals as Chordates.

Examples

Hemichordata or Stomochordata – *Balanoglossus* (acorn worm) and *Glossobalanus*.

Urochordata or Tunicata - *Ascidia*, *Ciona*, *Salpa* and *Doliolum*.

Table 6.1 Differences between Chordate and Non-chordate Animals

S.No.	Chordates	Non-chordates
1.	Notochord present	Notochord absent
2.	Central nervous system is dorsal, hollow and single	Central nervous system is ventral, solid and double
3.	Pharynx perforated by gill slits	Gill slits are absent
4.	Heart is ventral	Heart is dorsal
5.	A post-anal, metamerically segmented tail is present	Terminal part (pygidium) is unsegmented

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Cephalochordata or Acrania – *Branchiostoma* (Amphioxus or lancelet).

In the members of subphylum **Vertebrata**, the notochord is present during the embryonic period and replaced by vertebral column in the adult. A series of vertebrae surround the notochord along with dorsal nerve cord. Thus, all vertebrates are chordates but all chordates are not vertebrates.

Besides the three chordate characters, vertebrates have a ventral muscular heart with two, three or four chambers, kidneys for excretion and osmoregulation, and two pairs of lateral appendages, fins or limbs.

On the basis of absence or presence of jaw the sub-phylum Vertebrata is further subdivided into two sub groups, super class **Agnatha**, which lacks jaws, and super class **Gnathostomata**, which bears jaws. Only one class of living members, class Cyclostomata, represents the super class Agnatha. All other vertebrates which possess jaws are included in the super class Gnathostomata. They are divided into six classes namely Chondrichthyes (fish with cartilaginous skeleton), Osteichthyes (fish with bony skeleton), Amphibia (dual amphibious life), Reptilia (dry scale covered body), Aves (feathered body), and Mammalia (milk-producing gland). Animals of classes Cyclostomata, Chondrichthyes and Osteichthyes bear fins for locomotion. The animals of other four classes are four-limbed, called Tetrapods. These classes are Amphibia, Reptilia, Aves and Mammalia. In the subsequent text you will study salient features and general characters of different vertebrate classes.

Class Cyclostomata — All living members of the class Cyclostomata are parasites on some fishes. Cyclostomes have an elongated body bearing 6-14 pairs of gill slits in their gill pouch for respiration and containing a sucking and circular mouth. Their body is devoid of scales and paired fins. A single dorsal nostril leads into a closed nasal sac. A functional pineal eye is present just behind it. Single sex organ discharges gametes in the well-developed coelom. Cranium and vertebral column are cartilaginous with persistent notochord. Heart is surrounded by a cartilaginous capsule. Stomach is absent. Circulation is a typical vertebrate type. Lampreys are marine and migrate for spawning into rivers (Fig.6.19).

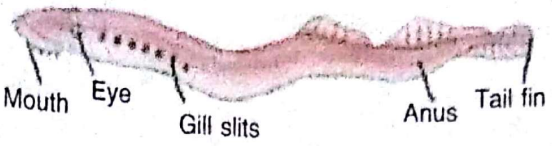

Fig. 6.19 *Jawless vertebrate- Agnatha (Lamprey).*

After spawning they stop feeding and within a few days they die. Their characteristic larvae after metamorphosis migrate to the ocean.

Examples

Petromyzon (Lamprey) and *Myxine* (Hag fish).

Class Chondrichthyes — Class Chondrichthyes includes all marine fishes with cartilaginous endoskeleton. There are about 600 species of cartilaginous fishes found all over the world. The body is streamlined with five pairs of gill slits without any gill cover. Gill slits may be present either on the lateral or ventral side of the head. The commonly known chondrichthyes are sharks, skates and rays (Fig. 6.20). Sharks are fast- swimming predators whereas the rays and skates are slow, bottom- living scavengers and mollusc-feeders. Sharks may vary in length. These may be 30 cm or may attain a length up to 16 m. Swimming is forbidden at some sea

(a)

(b)

Fig. 6.20 *Cartilaginous fish (a) Shark (b) Ray*

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Fig. 6.21 Bony fishes : (a) Flying fish (b) Flat fish (c) Sea horse (d) Eel (e) Angler fish

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beaches such as south-west pacific ocean because of danger of killer sharks. This has led to the establishment of elaborate protective measures, including aerial patrols, on eastern Australian beaches. The man-eating great white shark attains a length of more than 12 m. The jaws of shark are famous for very strong backwardly directed acrodont teeth (teeth attached to jaw bone). The skin is tough and covered with scales. Unlike bony fishes, sharks do not possess a swim bladder, the air sac, which, regulate buoyancy. Due to this reason sharks have to swim constantly else they will sink to the bottom. Sharks are viviparous and bring forth their young alive. The liver of shark is a rich source of vitamin A. Skates and rays are equipped with specialized organs. For example, *Torpedo* (electric ray) possesses electric organs as modified musculature between eyes and pectoral fins. Electric organs are capable of generating strong electric shock to paralyse the prey. *Trygon*, the stingray possesses a poison sting on its tail. The poison so released is strong enough to stun an animal.

Examples

Scoliodon (Dog fish), *Scyllium* (Shark), *Torpedo* (electric ray), *Trygon* (sting ray) and *Pristis* (Saw fish).

Class Osteichthyes — There are about 25,000 species in the class Osteichthyes. The characteristic feature of these fishes is bony endoskeleton, the skin is covered by cycloid scales. In contrast to cartilaginous fishes, bony fishes occur in fresh as well as marine waters, from the deep sea to tiny ponds. A variety of shapes and sizes is demonstrated by bony fishes (Fig.6.21). Because of their decorative shapes and colours, many freshwater bony fishes are maintained in aquaria in homes. Most fishes used as food are bony fishes. The common Indian fresh water fishes which are used as food are catla, rohu, mrigal, kalbasu, and the marine fishes used in food are pomfret, Bombay duck and Indian salmon. Bony fishes use pectoral, pelvic, dorsal, anal and caudal (tail fin) fins in swimming. In the flying fish *(Exocoetus)*, the pectoral fin is large and modified to use for gliding several metres in the air, as the fish leaps out of water. A swim bladder is present which helps in buoyancy. As a result, a bony fish can stay at a particular depth without expending energy in.

swimming, contrary to the cartilaginous fishes. Because of their great variety in shape and size it is difficult to give description of a bony fish using a single example. The general features including arrangement of fins and position of swim bladder in a common bony fish is shown in Fig. 6.22.

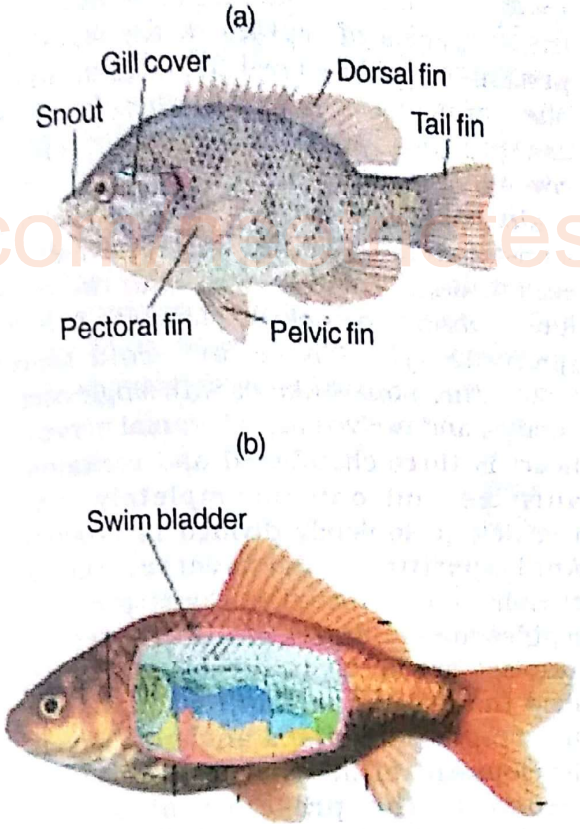

Fig. 6.22 (a) External features of a bony fish (b) Internal position of swim bladder

Fishes breed generally by laying eggs, and fertilization is external. A primitive form of nest building for laying eggs, and the care of eggs till hatching are exhibited by some species of fishes. In the sea horse, the males have a brood pouch, where eggs laid by the female, remain till they hatch.

Examples

Labeo (Rohu), *Catla* (Katla), *Lates* (Bhetki), *Puntius* (Punti fish), *Heteropneustis* (Singhi), *Clarius* (Magur), *Anabus* (Koi), *Channa* (Lata fish), *Exocoetus* (Flying fish), *Remora* (Sucker fish), *Echeneis* (Sucker fish), *Lophius* (Angler fish) and *Hippocampus* (Sea horse).

Class Amphibia — This includes about 3,000 species. As their name indicates, most amphibians have two phases in their lives. At the larval stage (tadpole) these are fish-like, swimming in water using a tail. After completion of larval stage they metamorphose into adults. Adults are morphologically different from their tadpoles. The adults live on land. For breeding, they require a watery environment because fertilisation is normally external. So amphibians, though generally adapted to life on land, have to be near water to complete their life cycle. They move using two pairs of limbs that contain four digits on the fore limb and five digits on the hind limb. Body is divided into head and trunk; there is no neck. The amphibian skin is moist and naked (without scales). The eyes have eyelids. A tympanum is present in place of external ear. Alimentary canal, urinary and reproductive tracts open into a common chamber, the cloaca, which opens to the exterior through an aperture called vent. The heart of frogs and toads is three-chambered. This is an advance over the two-chambered heart found in fishes. It receives blood separately from the lungs and the rest of the body, but pumps a mixture of both oxygenated and deoxygenated blood through a single ventricle.

These are cold-blooded (poikilothermous) animals with double occipital condyle and ten pairs of cranial nerves. Respiration in the tadpoles of frogs and toads and some urodeles take place by gills. After metamorphosis adults develop lungs which help in respiration. Some newts retain gills even in adult life. Sexes are separate, fertilisation is external and development through tadpole larva occurs in the aquatic environment. A common Indian toad is shown in Fig. 6.23.

***Fig. 6.23** Common Indian Toad*

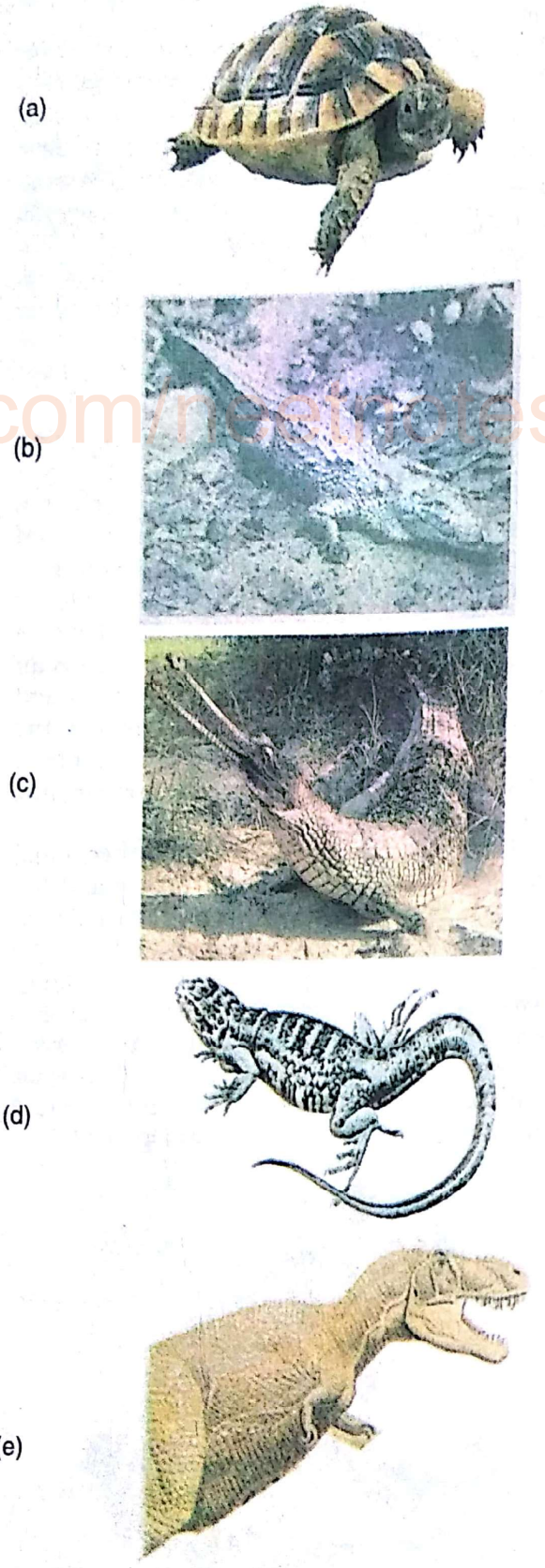

Fig. 6.24 *Reptiles (a) Common tortoise (b) Crocodile (c) Indian gharial (d) Monitor (Indian goh) (e) Tyranosaurus (extinct giant reptile)*

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Examples

Bufo (Toad), *Rana* (Frog), *Hyla* (Tree frog), *Rhacophorus* (Flying frog), *Alytes* (Midwife toad) *Necturus*, *Salamandra* (Salamander, *Ambystoma* (Salamander), *Trilototriton* (Indian salamander) and *Ichthyophis* (Limbless amphibian).

Class Reptilia — There are only about 6,000 living species of reptiles in the world. Most present-day forms are low statured animals, often crawling around and therefore named as reptile (Figs. 6.24, 6.25). Body of reptiles is covered by dry and cornified skin that contains epidermal scales or scutes and always contains a post anal tail. Reptiles do not have external ear openings. They can walk with two pairs of limbs, absent in snakes, each with five digits (pentadactyl). These are cold-blooded (poikilothermous) animals with single occipital condyle and twelve pairs of cranial nerves. The heart is three-chambered and contains two auricles and one incompletely divided ventricle (completely divided in crocodiles). Anal aperture is transverse, except in chelones and crocodiles. Most present-day reptiles are carnivores or insectivores. A few tortoises are herbivores. Snakes and lizards shed their scales as skin cast. Two features make reptiles truly land animals. One is the development of internal fertilization. The second is the presence of a special membrane, amnion, around the developing embryo.

Fig. 6.25 *Naja naja*

Examples

Chelone (Turtle), *Trionyx* (Turtle), *Testudo* (Tortoise), *Sphenodon* (Lizard - an example of Living fossil), *Hemidactylus* (House lizard), *Chameleon* (Tree lizard), *Calotes* (Garden lizard), *Draco* (Flying lizard), *Anguis* (Limbless lizard), *Phrynosoma* (Horned-toad), *Varanus* (Monitor), *Python* (Mayal), *Naja* (Cobra, Kautia), *Crocodilus* (Crocodile), *Alligator* (Alligator) and *Gavialis* (Gharial).

Class Aves — The class has about 9000 species of easily recognisable birds. Their characteristic features are the presence of feathers and the power of flight. There are very few birds, which have either wholly or partly lost the ability to fly. Some examples are emu, ostrich and cassowary. Birds have a reptilian ancestry. Birds possess scales on their hind limbs. Their eggs resemble reptilian eggs in general design with a calcareous shell. Indian sub-continent is rich in avian fauna. Some common Indian birds are shown in Fig. 6.26.

The forelimbs of birds are modified into wings (Fig. 6.27). The forelimbs contain three claw less digits whereas the hind limbs have four digits with claws. The hind limbs are modified for walking, swimming or clasping the tree branches. Presence of oil glands at the base of the tail and there is no other gland on the skin. The long bones of the endoskeleton are hollow and connected by air passages. This reduces the weight of the body. They have single occipital condyle and twelve pairs of cranial nerves. Birds are warm-blooded (homiothermous) and maintain a high metabolic rate and a constant body temperature. The heart is four-chambered. This

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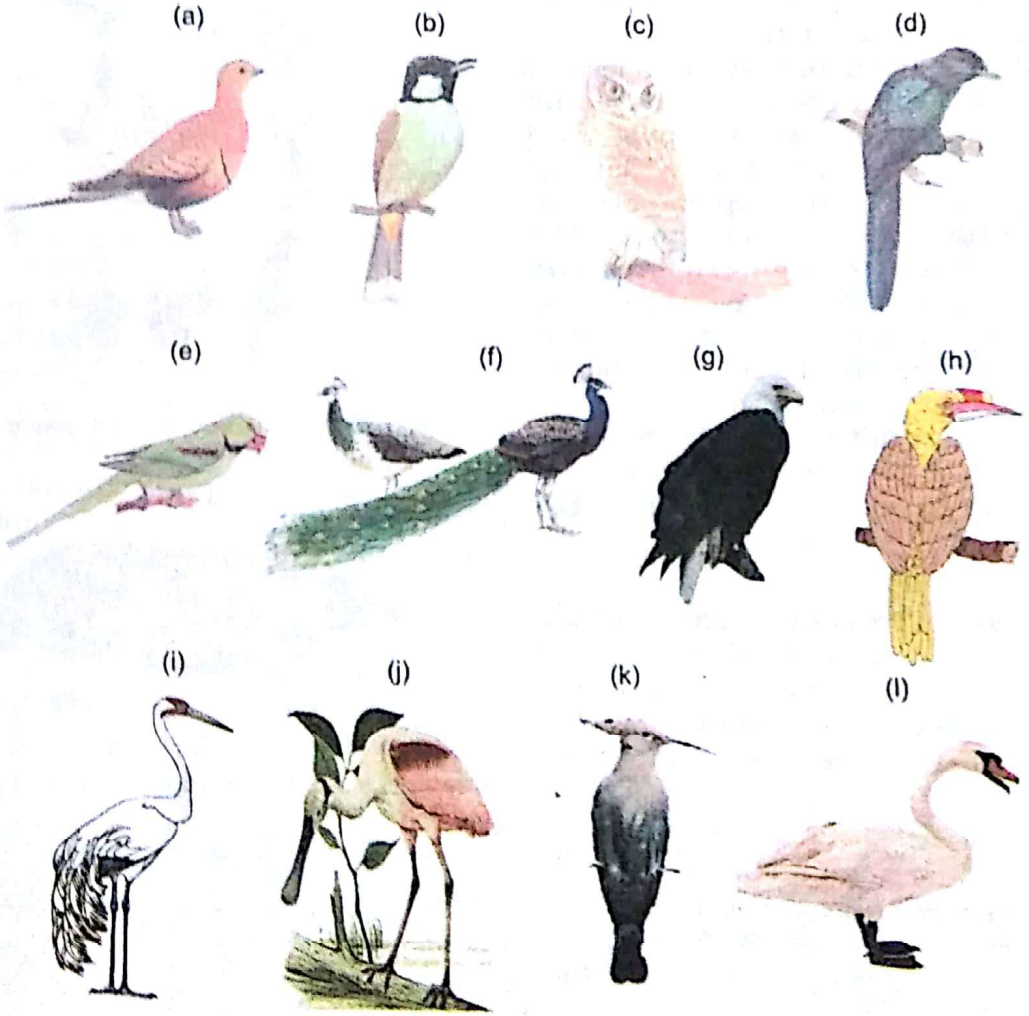

Fig. 6.26 Common Indian birds : (a) Pheasant (b) Bulbul (c) Owl (d) Koel (e) Parrot (f) Peafowl (g) Vulture (h) Hornbill (i) Crane (j) Spoonbill (k) Hoopoe (l) Swan

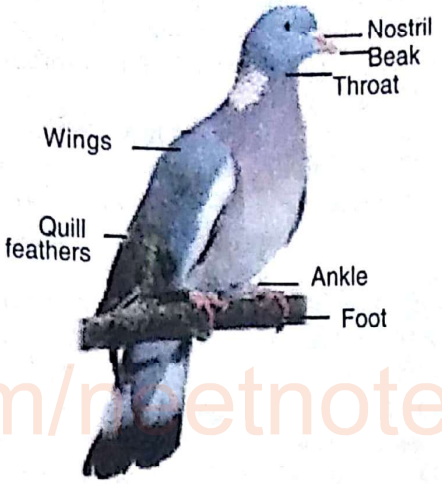

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Fig. 6.27 External features of a bird

allows complete separation of the oxygenated blood from the deoxygenated . Respiration is by lungs that contain air sacs.

Birds have no teeth; beak helps in feeding in many ways such as seed crushing, fruit scooping, flesh tearing, nectar sipping, wood chiseling and so on. The digestive tract has additional chambers, the crop and gizzard. The crop stores and softens food. The muscular gizzard helps in crushing and churning the food. Birds have good sense of sight. This is required to forage food, prey, landmarks and resting places from a height while flying. The brain is well-developed, particularly the regions concerned with the co-ordination of movement and balance. Birds lay large sized and yolky eggs (oviparous).

Examples

Ardea (Grey heron), *Corvus* (Crow), *Pavo* (Peafowl), *Gallus* (Fowl), *Alcedo* (King-fisher) *Columba* (Pigeon), *Psittacula* (Parrot), *Bubo* (Owl), *Phoenicopterus* (Flemingo), *Aqurilla* (Eagle), *Neophron* (Vulture), *Milvus* (Kites) and *Struthio* (Ostrich).

Class Mammalia — There are about 4,000 species of mammals found in the world. Mammals are perhaps the most successful and dominant animals today (Figs. 6.28-6.33). They thrive in variety of environments of the world – polar ice caps, deserts, oceans, mountains, forests, grasslands and dark-caves. The most unique mammalian characteristic is the milk-producing mammary

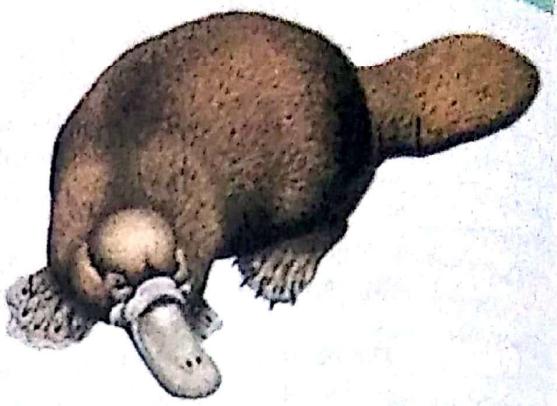
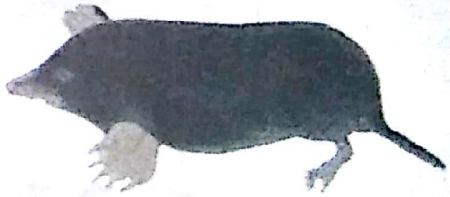


Fig. 6.28 Egg laying mammal - Platypus

Fig. 6.29 Kangaroo with its young in pouch

(a)



(b)

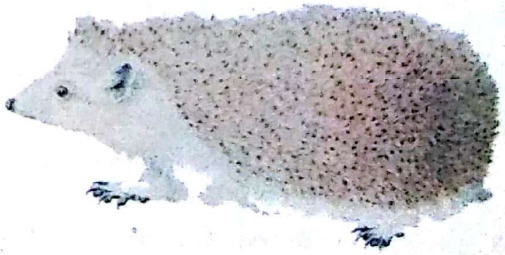


Fig. 6.30 Insectivores : (a) Mole (b) Hedgehog

Fig. 6.31 Flying mammal- Bat

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Fig. 6.32 Whale

glands by which the young ones are nourished. The skin of mammals is unique in possessing hair. External ears or pinnae are present in most mammals, which are not found in other groups of animals. Teeth are present in sockets. Such teeth are called thecodont. They are variously modified into incisors, canines, premolars and molars for the type of food ingested (heterodont). Mammals have a set of milk teeth, which are replaced by permanent teeth (diphyodont). A four-chambered heart ensures a continuous supply of oxygenated blood to all parts of the body. The lungs are well-developed. Breathing is enhanced by the muscular diaphragm, which separates the chest cavity from the abdomen. Mammals are generally terrestrial, found in a variety of habitats. Some mammals, however, have adapted to fly, for example bat (Fig.6.31), and live in water, for example whale (Fig 6.32).

The most advanced mammals are primates (Fig. 6.33). We, being the mammals, dominate the world. The reasons being the most developed ability of speech through language, opposable thumb and logical thinking.

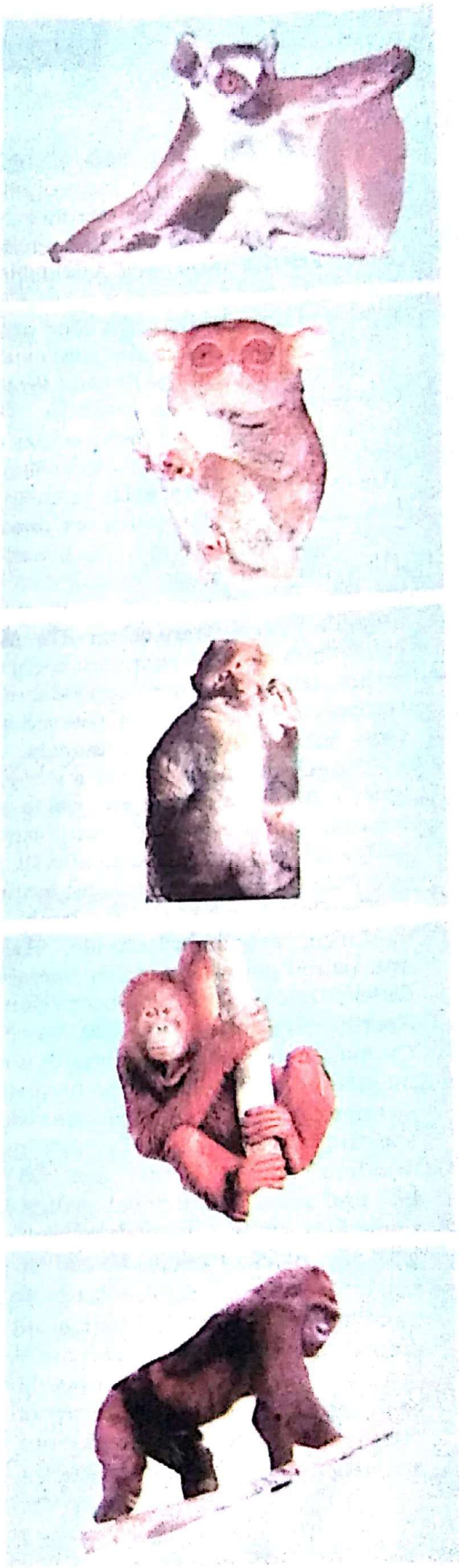

Fig. 6.33 Some primates (a) Lemur (b) Tarsier Simians (c) Monkey (d) Orang-utan (e) Gorilla (ape)

SUMMARY

Over a million different kinds of organisms have been reported and many more are yet to be discovered. The study of these organisms had been facilitated through classification which was based earlier on their morphology, and subsequently on natural relationships. Presently, evidences from comparative anatomy, embryology and palaeontology are being effectively used to derive taxonomic relationships. The heterotrophic mode of nutrition has been an important factor in causing great diversity and behavioural pattern. There are different grades of organisation and body plan, symmetry, body cavity, movement and reproduction are the features used in classification.

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The members of the Phylum Protozoa are acellular or unicellular animals which may be free-living, parasitic or symbiotic. These do not have distinct organs but cell organelles are present. Porifera has multicellular organisms without any tissue level of organisation but with characteristic flagellated collar cells. The body is perforated with numerous Ostia. The Cnidaria have special tentacles in the mouth and are mostly aquatic, being sessile or free-floating. Ctenophores are marine animals with transparent, flat or oval body shape. The Platyhelminthes exhibit a bilaterally symmetrical body which is flattened dorsoventrally. Their parasitic forms show distinct suckers and hooks. The Nemathelminthes includes parasitic as well as non-parasitic round worms. The Annelides are metamerically segmented animals with a true coelom. The Mollusca have a soft body surrounded by an external, calcareous shell. Respiration occurs in them by gills, lungs or through body surface. The arthropods have jointed appendages with an open type of blood vascular system, simple or compound eyes and body covered with the external skeleton made of chitin. This is the most successful group of animals.

The echinoderms possess a spiny skin with calcareous plates. These have long, movable spines, a mouth on the lower side and an anus on the upper side. Their most distinctive features are: presence of a water vascular system, excretion partly by diffusion through the body and partly by amoeboid cells in the coelomic fluid, occurrence of five pairs of sex organs, one pair per arm, and fertilisation in open water.

The Chordata includes the group of animals which possess a notochord either throughout or during early embryonic life. Their common features are a dorsal, hollow nerve chord and paired gill slits. Further, members of the Classes Cyclostomata, Chondrichthyes and Osteichthyes bear fins for locomotion. The members of remaining four classes, the Amphibia, Reptilia, Aves and Mammalia, move with the help of four limbs and are called tetrapoda. Cyclostomata, the most primitive notochords, are parasites on fishes. The Chondrichthyes include all marine fishes with cartilaginous endoskeleton. Of these, the sharks are fast swimming predators. These are viviparous and bring forth their young alive. The torpedo (electric ray) is capable of generating electric shock to paralyse the prey. The amphibians which are only freshwater have two phases in their lives. At the larval stage these are fish-like and swim in water using their tail, whereas, the adults live on land and move about using four limbs. The body of reptiles is covered by dry and cornified skin. They can walk with two pairs of limbs, absent in snakes, each with five digits. The two features which make reptiles truly land animals are: development of internal fertilisation and presence of amnion – a special membrane around the developing embryo. The birds are warm blooded animals with their fore-limbs modified into wings for flying and the hind limbs with four digits adapted for perching, walking or swimming. The mammals have a body divisible into head, neck, trunk and tail. Their other important features include body covered with hair, presence of external ear, presence of mammary glands, and the foetus (embryo) being nourished by the mother through a placenta.

EXERCISES

1. Fill in the blanks:
   (i) Variability of animal _____ has greatly helped in their classification.
   (ii) Heterotrophic mode of nutrition is an important factor in causing great diversity in the _____ of animal structure.
   (iii) Fertilisation results in the formation of _____ zygote.
   (iv) The digestive cavity of *Hydra* has only _____ opening for both ingestion and egestion.
   (v) Radial symmetry means that the body of the animal may be divided into _____ equal halves.

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2. Define metamerism.
3. Outline the role of coelom in animals.
4. The absence of locomotory organs in a number of protozoans has led them to acquire free-living and holotrophic mode of life. Comment.
5. Justify: "Conjugation is the first and clear evidence of sexual reproduction".
6. Discuss the various modes of asexual reproduction in animals.
7. Which of the following forms lining of canals of sponges:
   (a) Flagellated cells (b) Flattened cells
   (d) Both flagellated and flattened cells (d) None of the above.
8. The body wall of *Hydra* is:
   (a) Monoblastic (b) Diploblastic
   (c) Triploblastic (d) Polyblastic
9. The digestive tract of nematodes is differentiated into:
   (a) Mouth and intestine (b) Mouth and pharynx
   (c) Mouth, pharynx and intestine (d) Mouth, pharynx, intestine and anus
10. *Hirudinaria* and *Pheretima* are the representative of the phylum:
    (a) Cnidaria (b) Platyhelminthes
    (c) Annelida (d) Nemathelminthes
11. The insects are able to survive on land in almost any environment due to
    (a) Presence of a chitinous cuticle (b) A tough covering on the body
    (c) Impermeability to water (d) All the above features
12. Which of the following statements are true and which are false?
    (a) The millipedes possess 70 to 100 pairs of legs.
    (b) The body of the molluscs is hard but is covered by a soft shell.
    (c) The fertilization in majority of echinoderms occurs in open water.
    (d) The echinoderms do not show any fundamental similarities with chordates.

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UNIT THREE

Cell and Cell Division

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In the previous Unit you have acquired familiarity with the diversity of life, and have studied as to how to classify the organisms. You are also acquainted with various types of nomenclature. All organisms are made up of cells, and the cell, indeed is the unit of life. In order to examine the structure of the cell and its inclusions, certain tools and techniques are needed, which range from simple light microscope to electron microscope. Techniques, such as chromatography and electrophoresis are used to separate and isolate various macromolecules present in the cell. Some organisms exist as a single cell and, in others, cells come together and share various functions. The organisms thus acquire the status of multicellular organism, and the functions are performed by various types of tissues, exhibiting division of labour. The cells may have a very simple organisation (prokaryotic) or may show a high degree of differentiation (eukaryotic). All cells arise by the division of pre-existing cells. The type of division through which the chromosomes are duplicated and distributed equally to the daughter cells is called mitosis. In the other type of division, meiosis, the number of chromosomes and the genetic material is reduced by half to form gametes. In this Unit you will be acquainted with tools and techniques for the study of the cell, unicellularity and multicellularity, structure of the cell, its molecules and cell division.

CHAPTER 7

TOOLS AND TECHNIQUES

All organisms are made up of cells which are rather minute in size and so cannot be seen with the unaided eye. Therefore, a variety of tools and techniques are required to study the structure of the cell and its molecules. The first tool ever used to observe cells was a primitive microscope designed by Robert Hooke in 1665. Since then, rapid strides were made and several advanced techniques and equipments have been developed. This chapter will acquaint you with some of these.

7.1 MICROSCOPY

Microscopy means the use of microscopes for studying the cells. Microscope is an indispensable instrument for a biologist. The microscope used by Robert Hooke was very simple and consisted of only a combination of magnifying lenses fixed in a tube. This could be designated as a **simple microscope** (Fig. 7.1). The microscope generally used in a school biology laboratory is a **compound microscope** (Fig. 7.2). It has a combination of lenses which comprises concave-convex lenses. It enhances the magnifying power as well as the resolving power. It, therefore, becomes necessary to study some of the basic principles of microscopy on which their utility is primarily based.

A microscope can be compared with a human eye in its organisation and function. Both have lens systems and, in both the images of the object are formed. One of the most important principles of microscopy on which its utility is based, is getting a **magnified image** of the object.

Magnification and Resolving Power

Optical instruments are used to magnify an object, which means that with optical instruments the size of the retinal image of an object can be increased. The ratio of this magnified image to that image formed on the retina of an unaided eye is termed as **magnifying power** of that instrument and can be represented by the following equation:

$$\text{Magnification} = \frac{\text{Size of retinal image with the instrument}}{\text{Size of the retinal image with unaided normal eye}}$$

Fig. 7.1 *A primitive microscope used by Robert Hooke*

The top of the tubular body of the microscope houses the ocular lens, and the other end of the tube, close to the object to be examined, has an objective lens. Both of these lenses have magnifying power. How much has the object (specimen being examined) been magnified can be obtained by multiplying the magnifying power of the ocular and objective lenses. For example, with an ocular lens of 10× magnification and the objective of 40×, the object will be magnified 400 times.

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Resolving power refers to the ability of a magnifying instrument to separate details of two closely placed objects. This capacity of the instrument has nothing to do with the magnification. In a given system, the image formed may not be magnified at all but may be resolved into details. Likewise, in another system, the image formed may be magnified but with no further details. The resolving power of an instrument or lens may therefore, be defined as the smallest distance between the two objects which can be identified as separate images with the aided eye. In other words, *resolution is the ability of a lens to separate or distinguish between small objects that are close together.* The resolving power of the unaided human eye is 100 micrometre.

Much of the optical theory underlying the designing of microscope was developed by German physicist Ernst Abbe in 1876. The minimum distance between the objects that reveals them as separate entities can be given by the **Abbe Equation:**

$$L_m = \frac{0.61\lambda}{NA}$$

In the above equation L_m, represents the limit of resolution. The value 0.61 is derived from the computation of a number of complex trigonometric ratios. **Numerical aperture (NA)** = n Sinα, where n, is the **refractive index** of the medium, and sin α is the sine of the half angle of light entering the objective lens from the specimen (Fig. 7.2). λ is the wavelength of the light used to illuminate the object. As per equation, if L_m becomes smaller, the resolution increases and thus the finer details can be discerned in a specimen. Remember that the resolving power is inversely related to the wavelength of light. Therefore, the highest resolution is obtained with the light

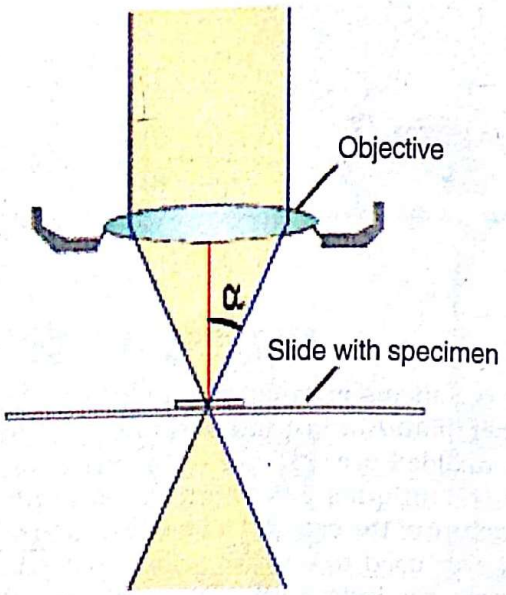

Fig. 7.2 *Semiangle of aperture*

of shortest wavelength i.e., (450 – 500 nm) light at the blue end of the visible spectrum. It is, therefore, customary to use blue filter in a microscope. In a common microscope, the light that strikes the specimen after passing through the condenser is cone-shaped. When this cone has a narrow angle and tapers to a sharp point, it does not spread out much after leaving the slide and does not form separate images causing low resolution. If the cone of light has a very wide angle and spreads out rapidly after passing through a specimen, closely packed objects appear widely separated and thus resolved.

The angle of light that can enter a lens depends on the refractive index of the medium as well as upon the object itself. The refractive index of air is 1.00. Since sin α cannot be higher than 1 (α 90° = 1), no lens working in air can have a numerical aperture more than 1. In fact, the angular aperture for the best objective lens is 70° (sin70° = 0.94). Therefore, the only practical way to increase the numerical aperture above 1.00 is to increase the refractive index by using an oil which has a refractive index identical to that of a glass. By replacing air with immersion oil, the light rays that could not enter the lens due to refraction or reflection will do so now.

The resolving power of microscope L_m also depends upon the numerical aperture of the

condenser as is evident from the following equation:

$$L_m = \frac{\lambda}{\text{NA Objective} + \text{NA Condenser}}$$

Most oil immersion objectives have a maximum numerical aperture of 1.4, with the numerical apertures commonly ranging from 1.0 to 1.35. However, it will be less than one if oil immersion is not used. This limits the overall resolution of the microscope. The maximum resolution of the light microscope, under the best obtainable conditions with light of the shortest visible wavelengths (approximately 426 nm), approaches 200 nm. In other words, two adjacent points closer together than 200 nm cannot be resolved into two separate images. Another important principle in choosing microscopic objectives is the **working distance**. The working distance of an object can be defined as the distance between the front surface of the lens and the surface of the cover glass (if used) or object when it is in sharp focus. The objective lenses of microscope with large numerical aperture and great resolving power have short working distances.

Light Microscope (Compound Microscope)

The light microscope consists of a sturdy strong metal body or a stand composed of a base and arm or limb to which the remaining parts are attached. A light source with a mirror or an electric illuminator is located at the base. Two focusing knobs are present on the arm, one of which is fine adjustment knob (smaller) and the other is coarse adjustment knob (larger). These knobs can move either the stage or the body tube (depending upon the make of the microscope) to focus the object (Fig. 7.3).

The stage is positioned half way up the arm and is used for holding the microscope slides by either simple side clips or by a mechanical stage clip. The mechanical stage clip is a device that allows the worker to move a slide smoothly by using stage knobs during scanning. The sub-stage **condenser** is mounted either within or beneath the stage. It helps to focus a cone of light on the object present on the slide. In a simple microscope, its position is fixed but in an advanced microscope it can be moved vertically. It is provided with a diaphragm so as to control the amount of light incident on the condenser lens.

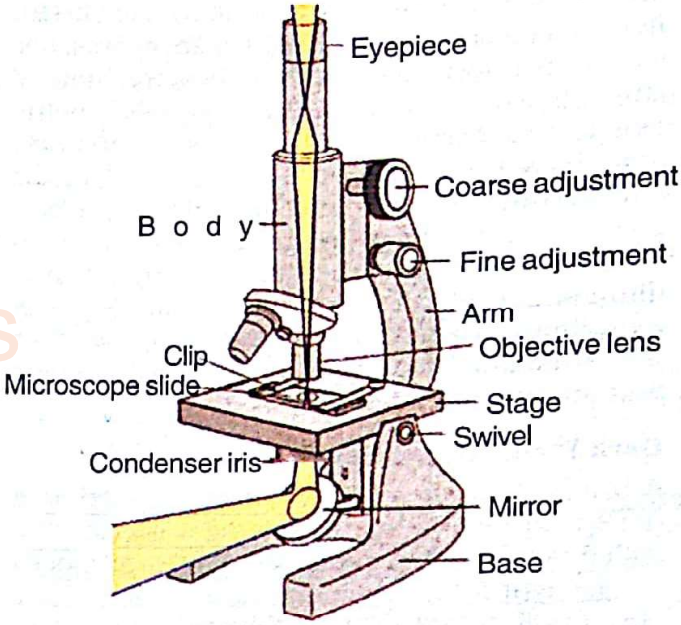

Fig. 7.3 *A compound microscope*

The curved upper part of the arm holds the body assembly of the microscope. The body tube towards the stage has a nose piece with one or more (usually 3 or 4) objectives mounted on it. The other side of the body assembly holds one or two **eyepieces** or oculars. The nose piece can be revolved in such a manner that one of the **objectives**, is directly in line with the eyepieces. In advanced microscopes, there are two eyepieces mounted for use by both the eyes. In such cases the microscope head is known as the **binocular head.** This assembly has a number of mirrors and prisms.

The objective lenses mounted on the nose piece are of different magnifying powers. These can be rotated along with nose piece to position any objective lens beneath the body tube. Ideally a microscope should be par focal. It means that the object should remain in focus when objectives are changed.

7.2 VARIATIONS IN THE OPTICAL MICROSCOPE

The above stated construction of a light microscope is for a **bright field microscope.** With special adaptations in lenses and condensers as well as

the light sources, three special types of microscopes can be described apart from bright field microscope. In a bright field microscope the image is formed when the light is transmitted through the specimen.The specimen, being denser and more opaque than its surroundings, absorbs some of this light and the rest of the light is transmitted directly up into the eyepiece. As a result, the image produced by the specimen will be darker than the surrounding brightly illuminated field. The bright field microscope is a multipurpose instrument and can be used for live, unstained materials as well as preserved and stained materials.

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Dark Field Microscopy

A bright field microscope can be adapted as a dark field microscope by adding a special disc called 'stop' to the condenser. The 'stop' blocks all the light from the central field, and now the object is illuminated by the oblique beam of light. This light is reflected off from the sides of the specimen itself. The resulting image is illuminated brightly against the dark background. The most effective advantage of this type of microscope is that, the nuclei, mitochondria and vacuoles of cells are readily detected.

Phase Contrast Microscopy

In this kind of microscopy, the property of internal structures having different densities is exploited as these can alter the light that passes through them in a specific manner. The microscope contains a phase shifting device that introduces some changes in light rays passing through the specimen resulting into differences in light intensity. The denser parts such as organelles alter the path of light more than the less dense cytoplasmic region. The image formed, therefore, has a varying contrast for different regions. The phase contrast microscope is thus most useful for viewing the living cells and for studying the internal structures of the cells such as the behaviour of chromosomes during mitosis or meiosis.

Differential Interference Contrast Microscopy

This type of microscopy is useful for studying unstained living cells. In these microscopes, the light rays are split into two beams before passing through the object by having additional refinements including two prisms that create contrasting colour in a image and two sources of light instead of one. One beam passes through the object and undergoes a phase change (diffracted wave). The second beam does not pass through the object and remains unchanged (undeviated wave). Since the two beams of light interfere with each other and also in a combined state, it is called an interference contrast microscope. By using this microscope, it is possible to find out the dry weight of macromolecules like DNA, RNA and proteins.

Fluorescence Microscopy

It is a specialised microscope in which UV radiations are employed (instead of visible light source) and is fitted with the complementary filters to protect the eyes of the viewer. The term microscopy originates from the fluorescence capacity of certain dyes (like acridine orange, auramineo and coriphosphine) and minerals when bombarded with UV radiations. The object to be viewed is coated with fluorochrome dyes. By illuminating the object with UV radiations the stained specimen will emit its own fluorescent wave-length (red, orange, yellow or green against dark field). This type of microscopy has wide applications in diagnosis of infections caused by bacteria, protozoans and viruses. The **immuno-fluorescent-antibody-labelling** technique involves conjugating the antibodies with fluorochromes by using two or more different antibodies linked to two or more such dyes. Thus, the distribution of two or more specific molecules within a cell can be studied.

Electron Microscopy

In principle, the electron microscope (Fig. 7.4) is a derivative of the compound microscope and employs the analogous components but not necessarily the same. In this method, the image is formed with a beam of electrons (instead of light) that can be made to travel in a wave-like pattern when accelerated to high speed. Waves are 1,00,000 times shorter than the waves of visible light. Because of this character the resolving power of an electron microscope is very high. The magnification of image is brought about by using two kinds of electromagnetic lenses (in place of glass lenses of light

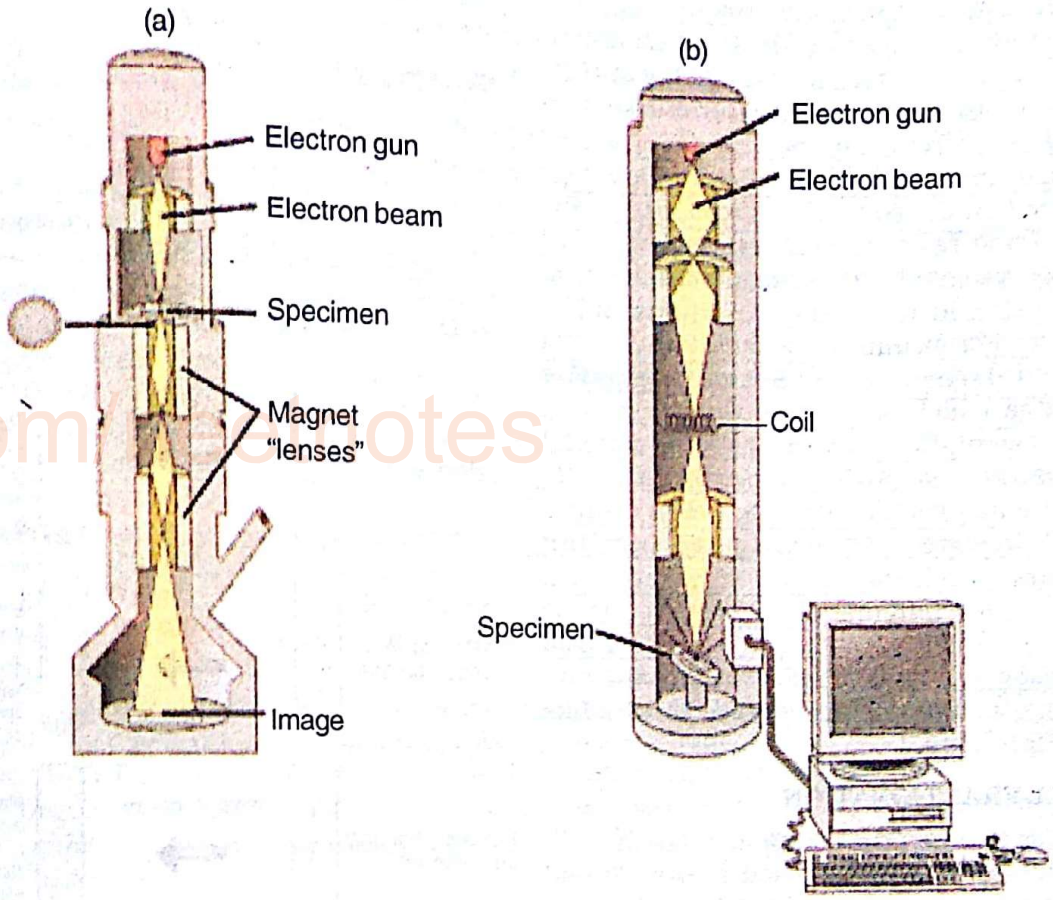

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Fig. 7.4 *Schematic diagram of (a) Transmission electron microscope (b) Scanning electron microscope*

microscopy). It has a condensing lens, a specimen holder and a focussing apparatus (objective lens and magnetic lens). The latter lenses are for magnification of the image. The path of electron in electron microscopes is controlled by electromagnetic lens like the path of light in light microscope through objective and ocular lenses. As the electrons are being used to create the image, the path and movement of electrons are established in high vacuum. The enlarged image is produced on a viewing fluorescent screen rather than being observed through the eyepiece.

The specimens for study in electron microscope are treated with chemicals or dyes to enhance the contrast and as such the live objects cannot be observed. Since images produced by electrons do not have the colour, the electron micrographs always have shades of black, grey and white. The colour enhanced micrographs are produced by computer aided shading.

There are two general types of electron microscopes, (a) Transmission electron microscope and, (b) Scanning electron microscope.

In the **transmission electron microscope** the electrons pass through the object and produce an image. The specimen, therefore, is cut into ultra thin sections. These sections are then treated for enhancing contrast by staining (coating) with the salts of various heavy metals (e.g. lead, tungsten, uranium). This process is necessary because most of the constituent elements in biological materials are of low mass and hence the contrast is weak. The coating also helps the material to withstand the electron bombardment. The specimen is placed on a copper grid to allow electron transparency to the specimen holder (Fig. 7.4a).

In **scanning electron microscope**, on the other hand, the image is produced due to reflection of electrons from the surface of the

specimen. The specimens, therefore, are dried properly and are coated or shaded with metals (gold, platinum) for creating a reflecting surface for incident electrons. The shower of electrons deflected back from the specimen are sensed by sophisticated detector and finally the image is displayed on the computer screen (Fig. 7.4b).

There have been several improvements in technologies related with refinement of electron microscopes and to develop variations in the basic plan. For example, most close relative of electron microscope is **Scanning probe microscope** which is capable of resolving to a very high extent the outer texture of the material and magnify it to 100 million times. This microscope has potential to image even a single atom. Likewise, **Scanning tunnelling microscope** scans the surface of the electrical conductors and computer chips for detecting the defects. Another variation is **atomic force microscope** which is useful in viewing the detailed pattern of the biological molecules like DNA and proteins.

7.3 CELL FRACTIONATION

With a view to study the components of the cell, the cell has to be **homogenised** or broken and components separated individually by different methods. The whole exercise is termed as **cell fractionation** (Fig 7.5). This involves homogenisation of the cell which can be brought about by (a) simple grinding of cells with glass beads, (b) breaking the cell by the physical forces like pressure, (c) using osmotic shock, and (d) employing ultrasound waves (Fig. 7.5A).

The prepared slurry is known as **cell homogenate**. Usually the intact cells or other debris may also be present along with the homogenate. These components can be removed from the homogenate by low speed **centrifugation**. The supernatant thus obtained is called cell free extract and is used to isolate further the different components like the various cell organelles, by different types of centrifugation.

Centrifugation

For fractionation of cell components from the homogenate, usually differential centrifugation technique is applied (Fig 7.5B). Centrifuge is an instrument that is used to create a centrifugal force on different components of cell. As the components are put to centrifugal force, they tend to move outward depending upon their size and density. The components, therefore, move in the medium with different speeds. This character and principle is exploited for separation.

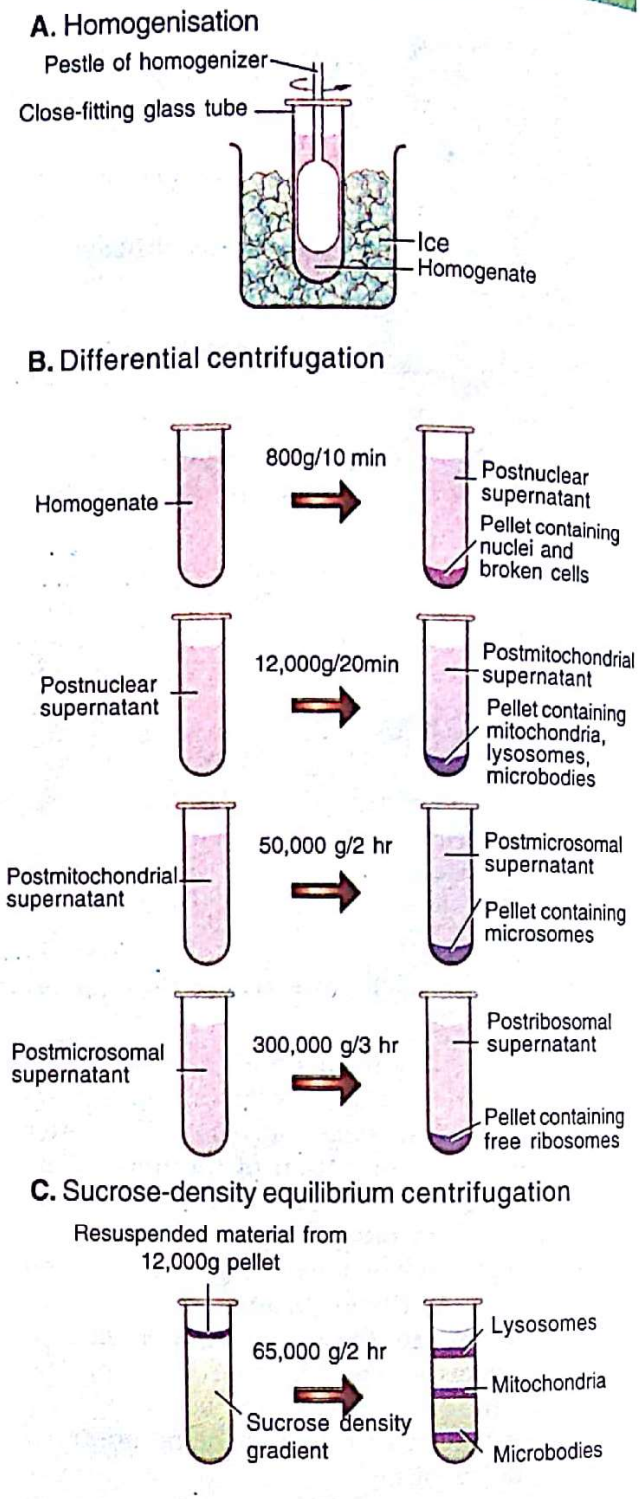

Fig. 7.5 *Steps in cell fractionation*

There are different kinds of centrifuges. Normal clinical centrifuge is a motorised instrument which can rotate upto 5,000 rpm (revolutions per minute) . The high speed centrifuge may have a speed upto 50,000 to 1,00,000 rpm and may or may not be refrigerated (refrigeration unit is meant to cool the chamber having the rotor or rotating unit). The high speed centrifuges are also called **ultracentrifuges**. The special feature in these centrifuges is that the chamber lodging the rotor on the drive apart from being refrigerated is also evacuated so that the rotor does not meet any resistance while revolving. The centrifugal force in such a centrifuge can reach about 5,00,000 times that of the gravitational force.

Depending upon the specific requirement, different kinds of rotors are used to hold the cell homogenate/solutions to be fractionated during centrifugation. In most of the clinical centrifuges there are only swing-out rotors having suspended test tubes (Fig. 7.6). When a centrifuge is switched on, the suspended tubes spread out into horizontal position and begin moving. Thus, a good centrifugal force is exerted on the particles in the homogenate within the centrifuge tube.

Density Gradient Centrifugation

Usually in such cases centrifugation through a gradient of increasing sucrose density is performed (Fig 7.5C). In this, the denser solution is at the bottom and its density gradually decreases towards the top. Once the gradient is formed, the homogenate is layered at the top and centrifuged. After centrifugation, components from the homogenate with different moving speeds reach equilibrium with the gradient or may form bands at different positions (Fig 7.5C). Hence this type of separation is also called **isopycknic (equal density) centrifugation.** The rate of sedimentation of the components depends on the size of the component and is described as sedimentation coefficient or **Svedberg units**. On the basis of the knowledge of the sedimentation coefficient, the size and composition of the cell, and types of components can be determined. Thus, peroxisomes may be separated from lysosomes or fragments of agranular endoplasmic reticulum from granular endoplasmic reticulum.

Buoyant Density Centrifugation

In yet another modification, the molecules can be separated on the basis of their buoyant density independent of their size and shape. In this case a very high concentration of solution of sucrose or cesium chloride (CsCl) is used in the form of a very steep density gradient. In fact, during the course of prolonged centrifugation the molecules of sucrose/caesium chloride tend to form a continuous density gradient. During centrifugation, each component of the homogenate moves with different speed (depending on the size and shape), but each component moves down and reaches a position

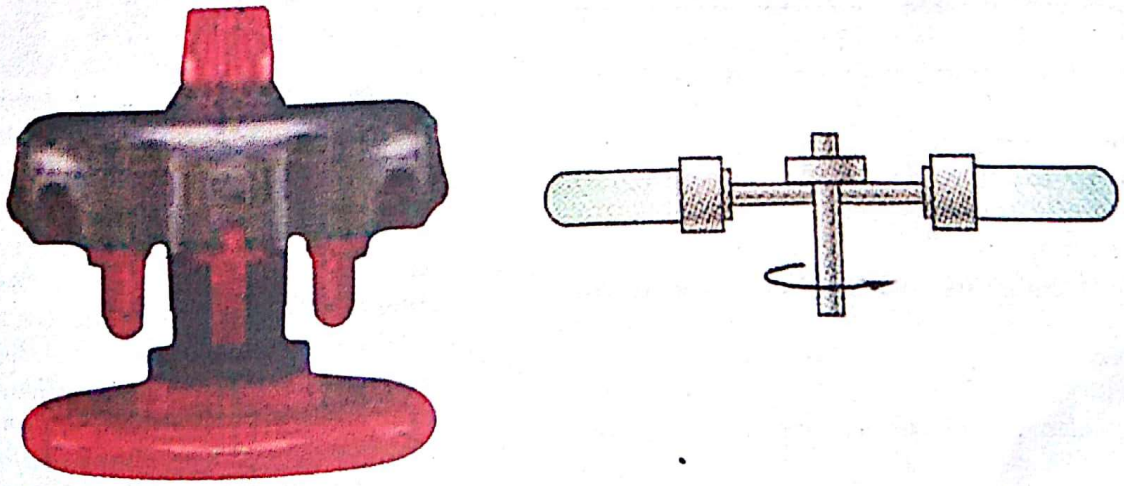

Fig. 7.6 *Centrifuge with swing-out rotors (a) at rest (b) tubes are in horizontal position during motion*

where the density of the suspension is equal to the buoyant density of a macromolecule. At this point the component floats and does not move any further due to its buoyancy. Thus, a series of distinct bands are formed not due to 'S' value (Sedimentation coefficient) but according to buoyant density, which is highest at the bottom and lowest at the top. This process of fractionation through centrifugation is also termed as **equilibrium sedimentation.** The method is so sensitive that it allows even separation of molecules having heavy isotopes (with ^{13}C or ^{15}N). The method was developed by Mathew Meselson and Franklin Stahl to separate heavy DNA with ^{15}N from DNA with ^{14}N, for providing evidence for semi-conservative replication of DNA.

Other Methods for Fractionation or Separation of Macromolecules of the Cells

There are a good number of methods, which can be employed for separation of molecules of the cells. However, the most important and commonly used ones are the following.

(a) Chromatography, and
(b) Electrophoresis

Chromatography

This is one of the most common methods of separation of molecular components of the cells present in a solution or **cytosol**, left after centrifugation. The cytosol is allowed to percolate in an insoluble medium, which has different affinity for molecules of different substances. In this technique, the principle of **partition coefficient** is exploited. The molecules migrate through the medium at different rates and are separated. At least, five kinds of chromatographic techniques have been developed. These are:

(i) Adsorption or column chromatography (commonly used for separation of mixture of tissue lipids),

(ii) Thin layer and paper chromatography (used for separation of amino acids, nucleotides, and low molecular weight products).

(iii) Ion exchange chromatography (used for insulin purification and plasma fractionation).

(iv) Gel filtration chromatography (used for determination of molecular weight of proteins).

(v) Affinity chromatography (used for separation of immunoglobulins, cellular enzymes and mRNA).

Electrophoresis

In this technique, the macromolecules (especially proteins, nucleotides, nucleic acid, etc.) are separated by differences in their net charge in the presence of externally applied electric field according to their molecular weight after nullifying their net charge under electric field (Fig. 7.7).

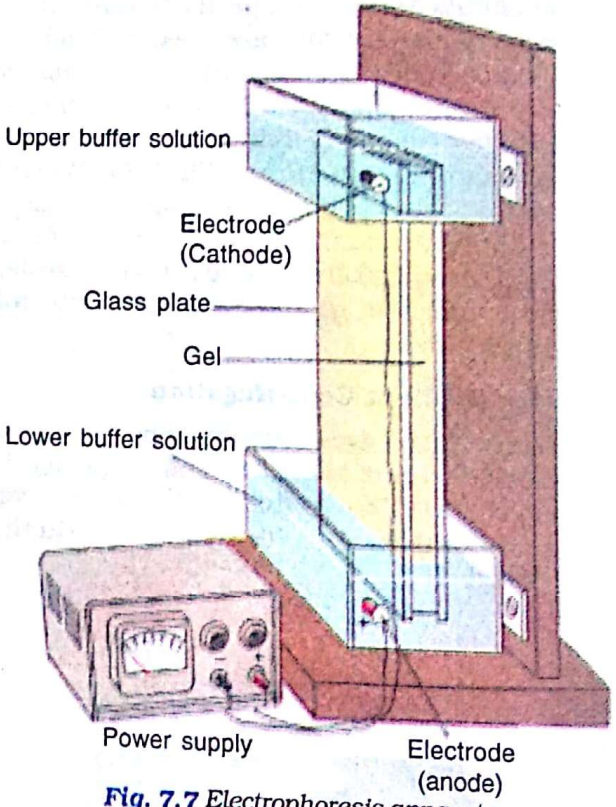

Fig. 7.7 *Electrophoresis apparatus*

This technique has also been exploited in a number of ways depending upon the requirement. For example, the base material through which molecules travel can be different: Polyacrylamide in polyacrylamide gel electrophoresis or PAGE, Agarose in agarose gel electrophoresis. The two dimensional-electrophoresis is performed in two directions. In one direction, the molecule may be separated

in non-denaturing conditions and in the second direction, at right angles to the first, under denaturing conditions.

Immuno-eletrophoresis is extremely sensitive and the molecule can be separated in picogram to nanogram quantities. Antibodies coupled with radioisotopes, specific enzymes or fluorescent dyes can be used for detection of specific proteins (even proteins that differ by a single amino acid can be resolved).

SUMMARY

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For studying the structure and function of cells, a number of tools and techniques have been developed during the past 300 years. The first and foremost is the microscopy. The light microscope is a simple microscope where two lenses are used. Compound microscope has three lenses. In the light microscope magnification power is :

$$\frac{\text{Sizeofretinalimagewiththeinstrument}}{\text{Sizeofretailimagewithunaidednormaleye}}$$

And the limit of resolution is given by the formula, $L_m = 0.61\ \lambda/NA$

A compound microscope is used for bright field microscopy, where the image is formed when light is transmitted through the specimen. The specimens absorb some of the light and the rest of the light is transmitted directly up into eyepiece through ocular. As a result we can see the object. The dark field microscopy can be carried out by adding a special disc 'stop' to the condenser. By this method we can visualise small cells like bacteria. Phase contrast microscope contains the device that transforms the subtle changes in light ray passing through the specimen into differences in light intensity, so image formed has variable contrasts for different regions.

In fluorescence microscopy, the source of light is UV radiation along with a filter to protect the eyes of the viewer. The object to be viewed is coated with a fluorescent dye. The stained specimen emits its own fluorescent wavelength when UV radiation is passed. In electron microscopy, a stream of electrons is used to view the object. The waves of electrons are much shorter than visible light. As such, the resolving power of microscope becomes very high. Only dead objects can be seen with it since they are treated with dye. The image is black and white, however, computer aided colouring can be accomplished. Electron microscopy is of two types: (i) Transmission electron microscopy in which the electrons pass through the object, and (ii) Scanning electron microscopy in which electrons are reflected from the specimen. Through this even the details of the pattern of DNA or protein can be viewed.

Cell fractionation is a technique to break cells into fragments and then study them. The cells are at first homogenised and then centrifuged. Subsequently, on the basis of sedimentation coefficient, size and composition of cell components is determined. In buoyant density centrifugation, centrifugation is done using high concentration of sucrose or cesium chloride solution. They form a continuous density gradient in the centrifuge tube and cell components settle in density, which is equal to their density.

Other methods include chromatography in which principle of partition coefficient is used. Chromatography is of different types – thin layer, paper, ion exchange, gel filtration and affinity. In electrophoresis, the cellular macromolecules are separated by differences in their net charge, when a electronic field is applied. The molecules can thus be separated even in pico- or nanograms. It is a very sensitive technique.

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CHAPTER 8

THE CELL : THE BASIC UNIT OF LIFE

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All organisms are composed of cells. All cells come from pre-existing cells. These two statements constitute the cell theory. The study of structure and composition of cells is known as cytology. Nowadays, attempts are being made to correlate the structure of cells with their function, including the life processes of the cells. This branch of biology, called Cell Biology combines many biochemical techniques and is concerned with the organisation and functioning of the individual cell. Cell biologists often deal with fundamental processes that are common to all cells. Hence, cell biology appears to be a unifying subject and studying cell biology is in some sense the same as studying life.

8.1 CELL AS A BASIC UNIT OF LIFE

We cannot imagine an organism that is not formed of a cell. Organisms may be made up of one or more cells. If the organisms are made up of a single cell, these are called unicellular organisms, e.g. *Amoeba*, *Chlamydomonas*, bacteria and many fungi. On the other hand, if organisms are made up of many cells, these are called multicellular organisms. The multicellular organisms may be made up of a few cells (e.g. some algal and fungal forms) to several million cells (e.g. human beings, a tree, whale, etc). In a multicellular organism certain cells become specialised to perform a specific function and thus division of labour is established among different groups of cells. The cells having a common origin and performing a similar but specific function constitute a **tissue** (e.g. muscle). Several types of tissues may join collectively to form an **organ** that carries out one or more specific functions (e.g. kidney, liver, leaf, and roots). In majority of animals, several organs are interrelated to perform a specific function and thus constitute an **organ-system** (e.g. digestive system, excretory system, etc.). Several types of organ-systems in the body of an organism show unique example of **division of labour**.

Whether plant or animal, the life of every organism begins as a single cell. The unicellular organisms complete their entire life cycle as a single cell. In others, an increase in the number of cells takes place in the course of life. All the cells of our body came from a single cell, the zygote, which divided continuously to form our multicellular body.

The cells are not only building blocks of the body, they are functional unit of life too. In the body of an organism a number of different types of cells may coexist. In fact, the activities of an organism are sum total of coordinated activities of its constituent cells. Cells in the body have the same genetic material, though mature cells may become specialised to perform a specific function. All the new cells of an organism develop from the pre-existing ones and hence, each cell has the same genetic information. It is therefore, capable of giving rise to a complete individual. This potential of the cell is termed as **totipotency**. Each cell is built up of several organelles and the cell performs all functions through its organelles present within the cytoplasm. Thus, all the activities of an organism are present in miniature form in each and every cell. Therefore the cell can be called a **functional unit of life**.

8.2 DISCOVERY OF CELL

Robert Hooke (1665) is credited with the discovery of cell. Hooke observed a honeycomb like pattern in a very thin slice of cork (Fig. 8.1). This honeycomb like structure consisted of a thick wall enclosing box-like compartments for which he coined the term cellulae for the first time and this term is synonymous to what we call cells. He regarded these cells (**cellulae**) as

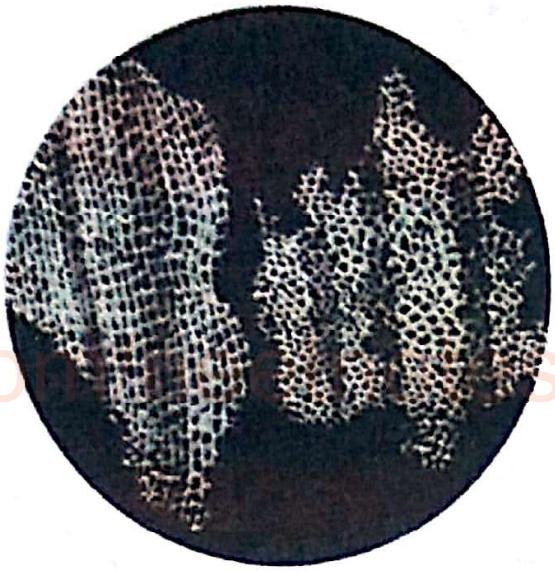

Fig. 8.1 *Box-like compartments in Cork tissue of plant*

passages for conducting fluids. Anton van Leeuwenhoek (1683) was first to observe free cells, like bacteria, protozoa, red blood cells and sperm, etc using a simple microscope. Alfonso Corti (1772) observed living substances in the cells.

An important discovery made by Robert Brown (1831) was the presence of a small sphere within the cells of *orchid* roots. This rounded body was later called **Nucleus** and was thought to be of common occurrence in the cells. Subsequently, Hugo yon Mohl (1838-1846) and Johannes Purkinje (1839) called the jelly-like substance **Protoplast**.

8.3 CELL THEORY

The interest of scientists in the cell structure and continuous refinement in microscope associated with improvement in techniques of observations resulted in accumulation of more knowledge about the cell. Matthias Schleiden (1838), a German botanist, examined a large number of plant tissues. He observed that all of them were composed of either one or the other kind of cells. This led him to conclude that all plant tissues are made up of cells. At the same time, Theodor Schwann (1839), a British zoologist, also studied different types of animal cells. Though he was able to observe the nuclei but he could not locate the cell wall. Instead these cells were having a thin outer layer, which we now know as **plasma membrane**. He also examined the plant tissues and realised that cell wall forms a unique character of plants. Yet inside, both the plant and animal cells had a similar organisation. Both kinds of cells had a nucleus that was surrounded by a clear substance. On the basis of these observations, Schwann proposed the hypothesis that *the bodies of animals and plants are composed of the cells and products of cells.*

Schleiden and Schwann later compared their observations and discussed Schwann's hypothesis. Their combined views led to the formulation of the Cell theory. This theory, however, did not explain as to how the new cells are formed? An important extension of Cell Theory — all living cells arise from pre-existing cells — took a few more years to appear. Rudolf Virchow (1855) was the first to explain that the cells divide and new cells are formed from the pre-existing cells *(Omnis cellula-e cellula)*. Later Nägali (1846) and Rudolf Virchow (1855) modified the hypothesis of Schleiden and Schwann to give the cell theory a final shape. However, it was Louis Pasteur (1862) whose experiment successfully established that life originates from pre-existing life.

The cell theory as we understand today has the following two tenets:

(i) All living organisms are composed of cells and their products.

(ii) All cells arise from pre-existing cells.

8.4 UNICELLULAR AND MULTICELLULAR ORGANISMS

Most cells are tiny and their volume ranges from 1 to 1000 μm^3. The eggs of some birds are quite big. A unicellular organism has to perform a large number of functions for its survival such as absorption of nutrients, exchange of gases with environment and metabolism. To carry out these functions, the cell has to be sufficiently large to accommodate a large number of organelles in it. Secondly, it has to increase its surface area (Fig. 8.2). As a cell increases in volume, its surface area also increases, but not to the same extent. The biological significance of this phenomenon lies in the fact that the volume determines the amount of chemical activity of cells per unit of time whereas the surface area determines the amount of absorption and the amount of release of waste products by the cells.

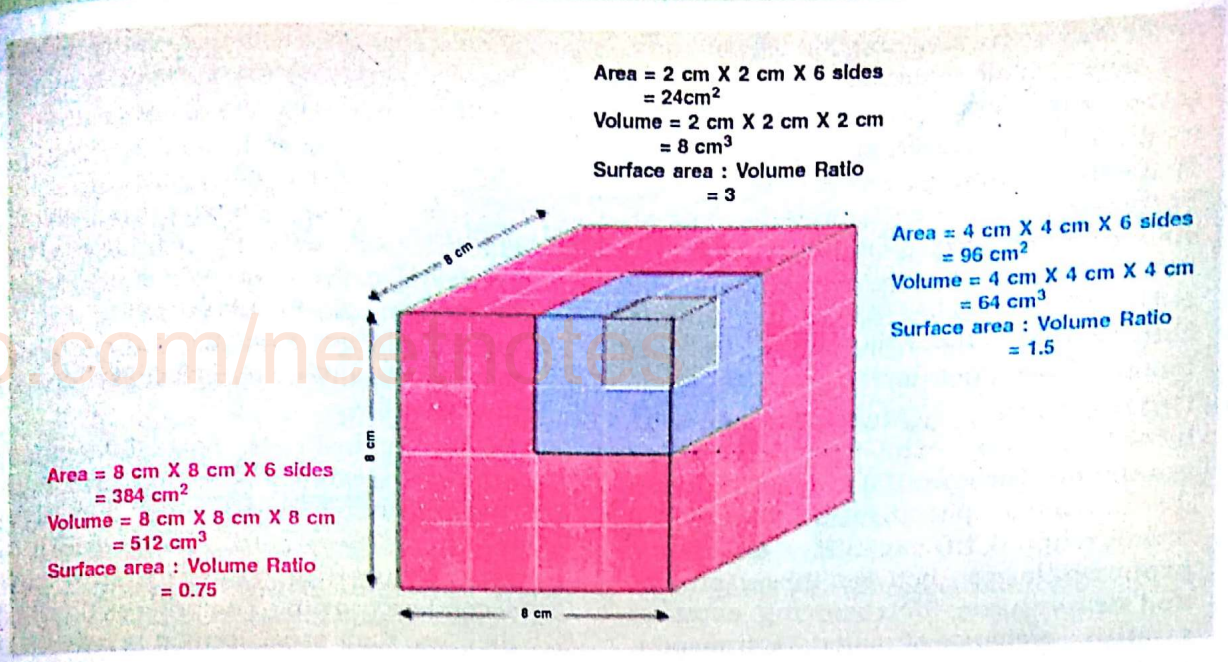

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Fig. 8.2 Surface area and volume ratio in a cell

As the living cell grows its rate of waste production and its need for intake of substances from outside increases at a rate faster than that of surface area. Small size of the cells of large organisms compensates disproportionate increase in the volume and surface area of cells. That is why, by and large, the cells are tiny; cells are small in volume to maintain a large surface area-to-volume ratio. Moreover, to maintain surface area-to-volume ratio in a balanced state, some cells have acquired additional structures in the form of projections such as **microvilli**. These increase the absorptive surface area.

Compared to a single-celled organism, a multicellular organism is not a mere aggregate of single cells. Rather, the cells undergo differentiation so as to share various functions which otherwise are independently performed by a unicellular organism. For example, some cells may secrete materials, which move outside the body for protective functions. Other cells may secrete substances, which hold the cells together. Still others may be differentiated to carry out absorption, photosynthesis, translocation of solutes, transmission of nerve impulse, and some cells get highly specialised to carry out reproduction. Even some dead cells have a role to play. For example, in animals the function of outer coat of dead cells forming a part of skin is to protect the inner living cells. In plants, conduction of water is performed by xylem vessels and tracheids, which are dead cells. Thus, a multicellular organism with its various types of cells is considerably more efficient than a single-celled organism. There may be other benefits that a multicellular organism is bestowed with or may have certain constraints in comparison to a unicellular organism. Some of the benefits and constraints of multicellular organisms are listed below:

(i) There is a unique co-ordination among cells of multicellular organisms like pumping of blood by heart muscle, and transmission of information (nerve impulse) through nerve cells.

(ii) In these capacities the cells have a dual existence as individual and as a part of the community (tissues).

(iii) Even if some cells die in a multicellular organism, the living cells can multiply and replace the lost cells, that is a clear cut benefit over unicellular organisms. For example, the cells of human skin, blood cells, etc.

(iv) The cells of multicellular organisms differentiate to various levels achieving a high degree of specialisation, even though they

are derived from a single zygotic cell and carry the same genetic material.

(v) Many of the differentiated cells lose one or the other basic activity, which is either suspended temporarily or irreversibly lost. For example, liver cells, muscle cells or epidermal cells retain mitotic activity after differentiation. The nerve cells and the red blood corpuscles (RBCs) lose mitotic activity after differentiation. RBCs even do not have a nucleus.

(vi) Differentiation bestows tremendous benefits on the multicellular organisms. These are: (a) increased survival (b) increased specialisation (c) ensured uninterrupted life activity, and (d) a proper balance between the cell surface and cell volume for receiving external stimuli, exchange of materials, transport, secretion, etc.

In general, the cells in an organism can be grouped under three major categories on the basis of the levels of differentiation:

(i) Undifferentiated cells: These cells are capable of undergoing division and development, for example, the **stem cells** (animals) and **meristematic cells** (plants).

(ii) Differentiated cells: These are post-mitotic cells which have undergone specialization or/and exhibit the division of labour. Therefore, these cells acquire a distinct character and perform a definite function. For example, RBCs carry out transportation of oxygen and carbon dioxide, the muscle cells perform kinetic functions or movement, and mesophyll cells carry out photosynthesis.

(iii) Dedifferentiated cells: Some differentiated cells are capable of reverting back to the undifferentiated meristematic state, when required. These cells are important for wound healing, regeneration, and secondary growth. The process by which they lose their specialization is referred to as dedifferentiation.

As you would have realised by now, cells of all organisms, whether unicellular or multicellular, have a close similarity in structure, molecular organisation and the various activities performed by them. This strongly suggests unity of life.

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SUMMARY

Cells were first discovered by Robert Hooke in 1665. Robert Brown discovered the nucleus in 1831. Schleiden and Schwann formulated cell theory, but did not explain how new cells are formed. Virchow (1855) explained that cells are formed from pre-existing cells. A unicellular organism carries out all life processes and activities within a single cell. Such organisms accommodate a large number of organelles by increasing the volume and surface area. On the other hand, in multicellular organisms there is a division of labour, and cells get differentiated to perform different functions. This enhances their survival abilities.

EXERCISES

1. Who discovered the cell?
2. Define totipotency.
3. What is meant by cell differentiation?
4. List the fundamental similarities in all cells?
5. Who proposed the cell theory?
6. Describe the cell theory.

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CHAPTER 9

STRUCTURE OF THE CELL

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You have learnt about the diversity of living world comprising organisms like microscopic bacteria to huge multicellular plants and animals. All are made up of cells which infact constitute basic unit of life. You have also studied in the previous chapter the different tools and techniques used for understanding structural aspects of the cells. In this chapter, you will be acquainted with structure and functions carried out by the various parts of the cell. It would enable you to understand the dynamics of tiny structure called cell.

In general, cells can be divided into two main types: (a) Prokaryotic cells, and (b) Eukaryotic cells. The prokaryotic cells have the simplest organisation, whereas the eukaryotic cells show a high degree of differentiation.

9.1 PROKARYOTIC CELL AND ITS ORGANISATION

The prokaryotic cells are represented by bacteria, blue-green algae, mycoplasma or PPLO (pleuro pneumonia-like organism), spirochete, and rickettsiae. Generally, prokaryotic cells are supposed to be far smaller than the eukaryotic cells. The prokaryotic cells, however, multiply more rapidly as compared to most of the eukaryotic cells. The prokaryotic cells have remarkable amount of variation in shape and size. There are four basic shapes of bacteria (Fig. 9.1):

(i) Bacillus: The most common shape is rod-like. The bacilli differ considerably in their shape of rod as it may be flat or round or cigar-shaped. The cells remain single but these may remain together after division to form pairs (*Diplobacillus*) or in groups of more than two (*Streptobacillus*);

(ii) Coccus or Spherical: The spherical cells may be present singly (*Monococcus*) or in groups of two (*Diplococcus*) or in long chain (*Streptococcus*);

(iii) Vibrios: A few rod-shaped bacteria may be slightly curved, or comma-shaped;

(iv) Spirilla: Many bacteria are shaped like a long, twisted spiral; these may resemble a comma. Other bacteria may be highly coiled and resemble a cork screw (*Spirochetes*).

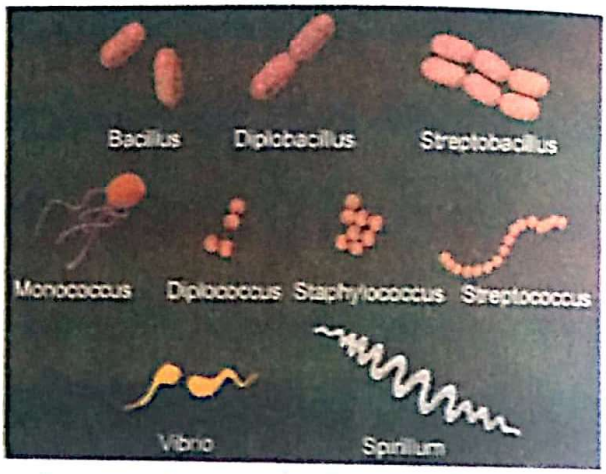

Fig. 9.1 Shapes of bacteria

Prokaryotic cells vary in size. The smallest bacterial cells are about 100–200 nm in diameter and approach a size very close to that of large viruses. However, most eubacterial cells are 1.1–1.5 µm wide and 2.0–2.6 µm long. A few bacteria and blue-green algae become very long in length (upto 500 µm) e.g. *Spirochetes* and *Oscillatoria*. A huge bacterium, *Epulopscium fishelsoni*, which was discovered in the intestine of the brown surgeon fish, is as large as 600 µm and as wide as 80 µm. It is now well established that a few prokaryotic cells are much larger than the normal eukaryotic cell.

A variety of structures is formed in prokaryotic cells (Fig 9.2). The structure and thickness of the wall itself differs in Gram-positive and Gram-negative cells. Despite these variations, prokaryotic cells are consistent in their fundamental structure and important

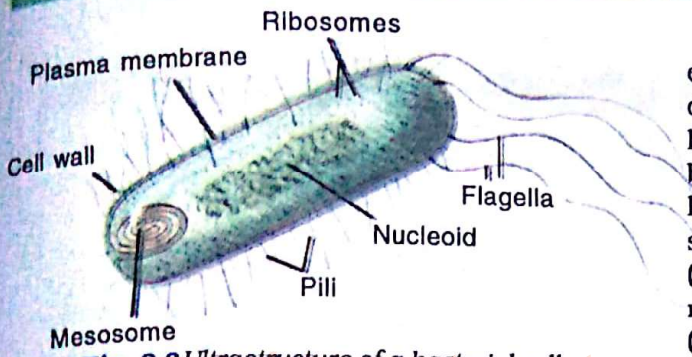

Fig. 9.2 Ultrastructure of a bacterial cell

components. Every prokaryotic cell is invariably bounded by a complex cell wall. Inside the cell wall is present periplasmic space, Beneath this lies the plasma membrane. The plasma membrane can be invaginated to form internal membrane structures. The prokaryotic cells do not have membrane-bound organelles. Hence, the interior appears very simple. The genetic material is localised within a discrete region, called nucleoid. The nucleoid is not separated from the surrounding cytoplasm by any delimiting membrane. The ribosomes and the inclusion bodies are randomly scattered in the cytoplasmic matrix.

Cell Envelope

Most of the prokaryotic cells, particularly bacterial cells, have a chemically complex cell envelope. The layers of cell envelope stack upon one another and are often bonded together tightly. In electron micrograph, three basic layers can be identified: the outermost **glycocalyx**, followed by **cell wall and cell membrane** (plasma membrane). Although each layer of the envelope performs a distinct function, together they act as a single protective unit.

Glycocalyx is the outermost layer comprising a coating of macromolecules, which protects cells and also helps in adhesion. This layer differs in thickness and chemical composition in different bacteria. Some have a loose sheath called **slime layer,** which protects the cells from loss of water and nutrients. Others may have a thick and tough covering known as **capsule.** The capsule and slime layer are made up of polysaccharides, but may sometimes contain proteins also. The capsule is responsible for giving gummy and sticky character to the cell. This layer is not absolutely essential for bacterial survival but can at times be highly specific and immunogenic.

Cell wall is the second layer of the cell envelope below the glycocalyx. This layer determines the shape of the cell and provides a kind of strong structural support to prevent a bacterium from bursting or collapsing in a hypotonic solution. This layer is rigid due to a special macromolecule called **peptidoglycan** (murein or mucopeptide). It is composed of repeating framework of long glycan strands (N-acetyl muramic acid and N-acetyl glucosamine), which are cross-linked by short peptide chains to provide a strong but flexible support framework. A number of antibiotics (e.g., penicillin and cephalosporins) inhibit cross linking of peptidoglycan strands. Therefore, cells undergo lysis in the presence of these antibiotics. **Lysozyme,** a naturally occurring enzyme in saliva and tears, also provides defense against certain bacteria by hydrolysing the peptidoglycan.

Gram staining is a special technique, which is used to classify bacteria into two groups, viz. Gram-positive and Gram-negative bacteria (Table 9.1). The bacteria are stained with weakly alkaline solution of crystal violet or gentian violet. The stained slide of bacteria is then treated with 0.5 per cent iodine solution. This is followed by washing with water and then with alcohol or acetone. There is difference between the cell envelopes of the two groups. In Gram-positive bacteria, the cell wall is thick and is primarily made up of peptidoglycan. In Gram-negative bacteria, the cell envelope shows three layers. The cell envelope is composed of outer membrane, and a thin layer of peptidoglycan followed by the plasma membrane. The Gram-positive cell wall is 20 – 80 nm in thickness and also contains tightly bound teichoic acids. The outer face of the outer membrane of Gram-negative bacteria contains lipopolysaccharides, a part of which is integrated into the membrane lipids. The inner face has a number of proteins which are anchored into peptidoglycan. The outer membrane of Gram-negative bacteria contains proteins called porins, and these proteins function as channels for the entry and exit of hydrophilic low molecular weight substances. In *Mycobacterium* and *Noccardia,* the wall is that of Gram-positive type but a part of their cell wall is made up of a very long chain of the fatty acid, called mycoic acid.

Table 9.1 Differences between Gram - positive and Gram - negative Bacteria

Gram- positive	Gram-negative
The bacteria remain coloured blue or purple with Gram staining even after washing with alcohol.	The bacteria do not retain the stain when washed with alcohol.
Outer membrane is absent.	Outer membrane is present.
Cell wall is 20-80 nm thick.	Cell wall is 8–12 nm thick.
The wall is smooth.	Wall is wavy and comes in contact with plasmalemma only at a few loci.
Murein content is 70 – 80 %.	Murein content is 10 – 20 %.
Basal body of the flagellum contains 2 rings.	Basal body of the flagellum has 4 rings.
Teichoic acid present in cell wall.	Teichoic acid absent in cell wall.
A few pathogenic bacteria belong to Gram-positive group.	Most of the pathogenic bacteria belong to Gram-negative group.

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Plasma membrane is an absolute requirement for all living organisms. In prokaryotic cells, the membrane forms the boundary of the cytoplasm being guarded from outside by extracellular matrix and the cell wall. The membrane is responsible for the relationship of a cell with the outside world. Another characteristic of the membrane is its semi-permeable nature. It is permeable to some substances to a variable degree and there are some carrier molecules embedded within the membrane, which bind to specific molecules and transport them in a specific direction. Therefore, the membrane controls flow of specific molecules into and out of the various compartments. Because of its quasi-fluid nature, it undergoes dynamic changes. The chemical composition of plasma membrane is variable, with lipids (20–79 per cent), proteins (20–70 per cent), oligosaccharides (1.5 per cent) and water (20 per cent) of its total weight, depending upon the tissue and the organism involved. The main lipid components of the membrane are phospholipids, glycolipids and cholesterol with their relative proportion varying in different cell membranes. Membrane associated lipids are asymmetric with polar and non-polar ends. These are also called **amphipathic** i.e., they contain both hydrophilic and hydrophobic regions (Fig. 9.3).

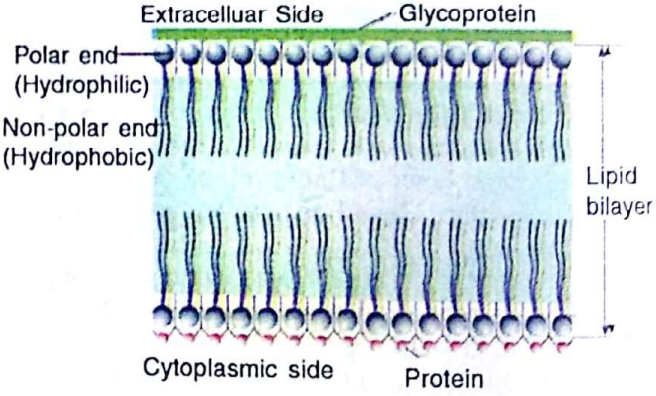

***Fig. 9.3** Unit membrane model*

The polar ends interact with water and are called hydrophilic, whereas non-polar ends are hydrophobic and these tend to associate with each other. The lipids form a bilayer and therefore, the outer surface of the membrane is hydrophilic. The hydrophobic ends are buried in the interior away from surrounding water and many of these amphipathic lipids are phospholipids. In bacteria, membranes are

different from those in eukaryotic cells in that they lack sterols such as cholesterol. The cell membrane is very thin, and through the freeze-etching technique it has been shown to have complex internal structure. There are small globular proteins that lie within the lipid bilayer. Some proteins are associated with the surface and are known as peripheral proteins. The major functions attributed to the membrane are performed by proteins. Like glycolipids, the proteins may also have oligosaccharide chains attached to them (glycoproteins) largely on the surface facing the exterior. These may contribute to formation of glycocalyx. The proteins in the membrane can be found in a variety of forms. These may be **transmembrane** proteins extending through the bilipid layer as a single helix (e.g., glycophorins). The **extrinsic proteins** on the other hand, may be present on the cytosolic face or towards the external face (e.g., **spectrin**). These extrinsic proteins may be covalently attached to fatty acid chains or noncovalently attached to other transmembrane proteins. Some of these proteins cannot be easily released in comparison to extrinsic proteins and so are called **intrinsic proteins**.

The universally accepted **fluid mosaic model** (Fig. 9.4) for plasma membrane is based on integration of data from chemical analysis and those from the study of biophysical properties with the help of a variety of techniques. This model was proposed by S. Jonathan Singer and Garth Nicholson (1972) and is the most acceptable one. According to these authors, the membrane is a continuous lipid bilayer having integrated protein molecules. The membranes are semifluid and dynamic in nature. The lipid and protein molecules help in performing

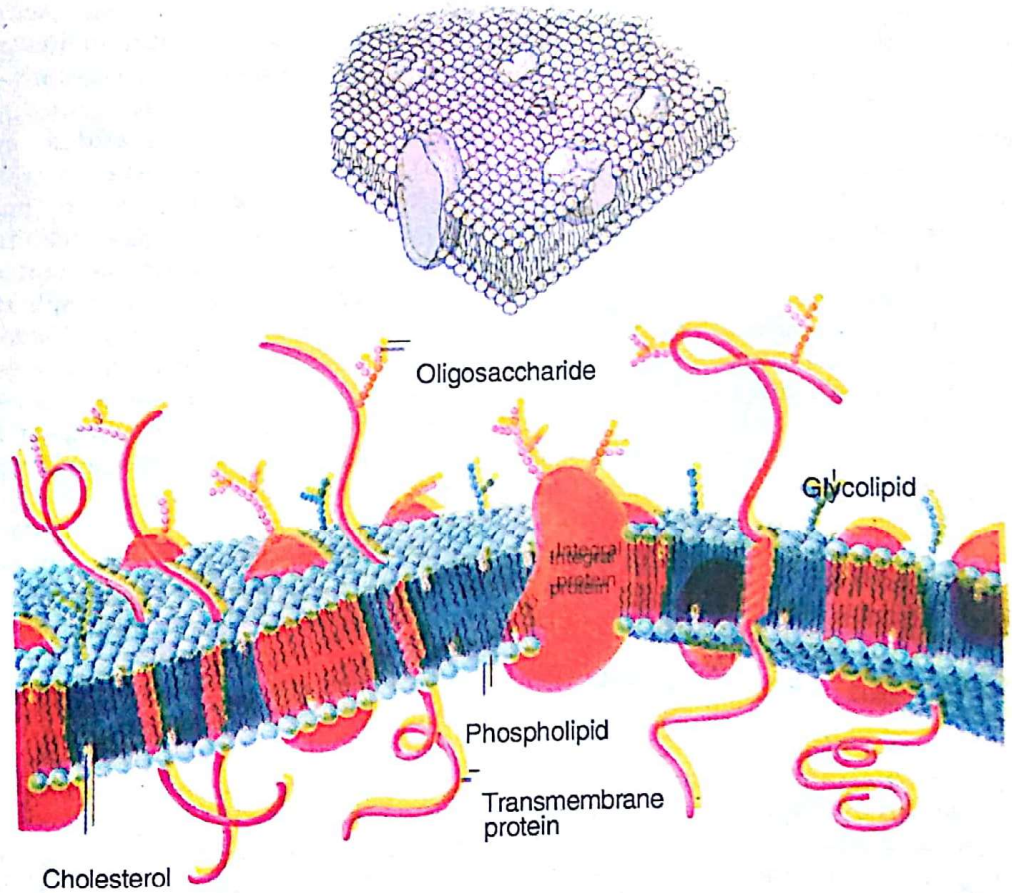

Fig. 9.4 *Fluid mosaic model of plasma membrane*

transport movements. Two types of proteins are distinguished. The peripheral proteins are loosely connected to membranes and, therefore, can easily be removed in aqueous solution. The rest of the proteins are integral proteins, which cannot be easily extracted. These proteins are insoluble in aqueous solution when freed from lipids. Some large integral proteins project beyond the lipid layer on both sides of the membrane and are considered as channel proteins through which the water soluble materials can pass. There are some integral proteins, which penetrate the lipid bilayer exposed to one surface only. Many integral proteins and some membrane lipids are associated with oligosaccharides; a part of these oligosaccharide chains project into the extracellular fluid. (Fig. 9.4)

The plasma membrane of bacterial cell must fulfill an incredible variety of roles successfully. It retains the cytoplasm, particularly in cells without cell wall and separates it from surroundings. It also serves as a selective permeability barrier allowing particular ions and molecules to pass either into or out of the cell while preventing others. It prevents loss of essential components through leakage and aids in movement of molecules, which otherwise will not cross the membranes. Such a transport system can be used for nutrient uptake, waste secretion, protein secretion, etc. Additionally, the bacterial plasma membrane is the location of critical metabolic processes like respiration, photosynthesis, synthesis of lipids and cell wall constituents. Finally, the membrane also holds receptor molecules that help bacteria detect and respond to chemicals in their surroundings. The prokaryotic cells do not contain complex membrane-bound organelles (like mitochondria, chloroplast etc.) but membranous structures of several kinds can be observed. One such common structure is **Mesosome**(See Fig 9.2). These are extensions of plasma membrane into the cell in the form of vesicles, tubules and lamellae. These are commonly seen in Gram-positive bacteria. These may be involved in: (a) cell wall formation, (b) chromosome replication and distribution to daughter cells, (c) respiration, (d) secretory processes, and (e) increase in plasma membrane surface area and enzymatic content.

Chromatophores are internal membrane systems in prokaryotic cells that may become extensive and complex in photosynthetic forms like cyanobacteria and purple-bacteria. In nitrifying bacteria, these membranes may form aggregates of spherical flattened or tubular vesicles, which assist in increasing the membrane surface for greater metabolic activity.

Inclusion Bodies

The reserve materials of bacteria are stored in the cytoplasm as inclusion bodies or storage granules. These are not bounded by any membrane system and lie free in the cytoplasm, e.g., **phosphate granules**, **cyanophycean granules** and **glycogen granules**. Some other inclusion bodies may be surrounded by a single layer non-unit membrane, which is 2–4 nm thick, e.g., poly-β-hydroxybutyrate granules, sulphur granules, and gas vacuoles.

Other remarkable organic inclusion bodies are the **gas vacuoles,** which usually occur in cyanobacteria, purple and green photosynthetic bacteria and a few other aquatic forms that are free-floating (planktonic). These are basically aggregates of a number of small, hollow cylindrical gas vesicles. Gas vesicles are not permeable to water but are permeable to atmospheric gases. Because of gas vacuoles, these bacteria keep floating on or near the surface of water. These vacuoles help the organisms in positioning themselves in the water column for trapping sunlight for photosynthesis, or for avoiding intense sunlight.

The two major inorganic inclusions are the polyphosphate granules or the **volutin granules** and **sulphur granules**. These granules take different colours with basic dyes, therefore, they are also termed as **metachromatic granules**. The volutin granules are phosphate polymers and function as a storage reservoir for phosphate. Some bacteria also store sulphur temporarily as sulphur granules. These may be formed when bacteria use hydrogen sulphide as electron donor during photosynthesis. These granules may accumulate in either periplasmic space or in special cytoplasmic globules.

The cytoplasmic matrix of the prokaryotic cell is often packed with ribosomes. Ribosomes are also associated with plasma membrane of the cell. They appear as small featureless particles at low magnification but in fact are very complex, both chemically and structurally. Ribosomes measure 14 – 15 nm by 20 nm. They are composed of two subunits – 50 S and 30 S, that make up 70 S for the complete ribosome (the heavier and the more compact the particle, the larger will be the Svedberg numbers). It is to be remembered that S-values are not proportionate to molecular weight, e.g., the sum of 30 S and 50 S subunits is 80 but the complete ribosome is only 70 S.

The ribosome is the site of protein synthesis. Matrix ribosomes synthesise proteins, which remain within the cells but the ribosomes on plasma membrane make proteins that are transported outside. Several ribosomes attached to a single messenger RNA (mRNA) form a complex known as polyribosome or polysome. The ribosomes in polysomes simultaneously translate the mRNA into proteins.

The prokaryotes do not have a membrane bound, well defined nucleus. Their genetic material (DNA) is neither complexed with other molecules (proteins) nor it is packed in the choromosomes. The genetic material in prokaryotes is composed of a single circular DNA, and is packed in a **nucleoid**. In some cases RNA and a small amount of protein are present . The length of the DNA strand may be about 250-700 times the length of the cell. It is, therefore, efficiently packed in the cell to fit within the nucleoid region. The DNA is extensively looped and coiled with the help of nucleoid proteins. These proteins are different from histone proteins that are present in eukaryotes. The nucleoid is usually associated with mesosomes. In isolated preparations, nucleoid has been found associated with plasma membrane. It has, therefore, been concluded that bacterial DNA is attached to cell membrane and the membrane may be involved in separation of duplicated DNA into daughter cells during division.

Flagellum

The electron microscopic studies have revealed that the bacterial flagellum (Fig. 9.5) is composed of three parts - **filament, hook** and **basal body**.

The filament is the longest and the most obvious portion of the flagellum. It extends from the cell surface to the top. It is a hollow rigid cylindrical structure made of the protein called **flagellin**. The protein molecules are arranged in a spiral manner in the filament. The filament measures about 20 nm in diameter and 1-70

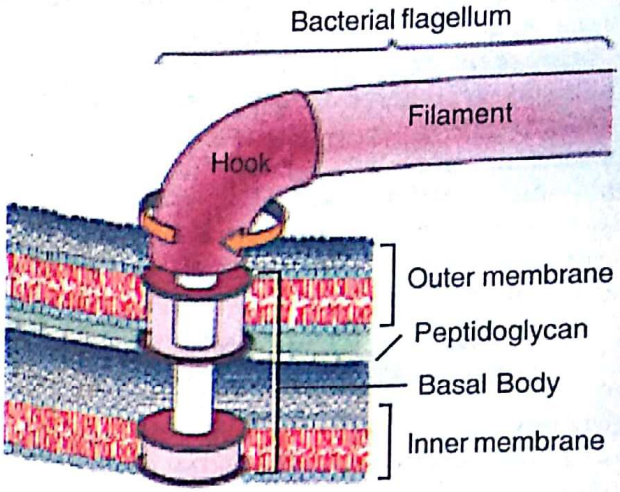

Fig. 9.5 *Basal structure of flagellum of bacteria*

nm in length. It is inserted into a curved tubular hook which is anchored to the cell by basal body. The hook is quite different from the filament and is made up of different protein subunits. The basal body is the most complex part of the flagellum and consists of four rings connected to a central rod in Gram-negative bacteria. The Gram-positive cells have only two basal body rings. The inner ring is connected to plasma membrane, while the outer ring is connected to peptidoglycan membrane. The filament, hook and basal body are arranged in such a manner that the structure permits the filament to rotate by 360° rather than undulating back and forth like a whip. As the flagellum rotates, it spins the body in the opposite direction and pushes the bacterium in forward direction.

Pilus and Fimbriae

These two terms have been used interchangeably to indicate any bacterial surface appendage not involved with motility. The **pili** (*sing.* pilus) are elongate, tubular structures (Fig. 9.6) made up of a special protein

called **pilin**. True pili have been reported only in Gram-negative bacteria so far and in these forms they are involved in mating process. During this process, usually partial transfer of DNA from one cell (**donor cell**) to another cell (**recipient cell**) takes place. Formation of pili is genetically controlled and is specific for a cell type as conjugation takes place between compatible bacterial cells. The **fimbriae** are small bristle-like fibres sprouting out of the cell. These seem to be slender tubes composed of helically arranged protein subunits, which are 3–10 nm in diameter. Some type of fimbriae are known to attach bacteria to solid surfaces such as rocks in streams and to host tissues. These are also responsible for mutual clinging of cells forming a film on the liquid as well as other thick aggregates.

9.2 EUKARYOTIC CELL AND ITS ORGANISATION

Eukaryotic cells, by definition, have a nucleus, which contains nuclear material enclosed by a double layered membrane. All plant and animal cells fall under this category. In general, the eukaryotic cells have the following components: cell wall (absent in animal cells and some protists), plasma membrane, cytoplasm and organelles. Until the discovery of electron microscope, the cytoplasm was thought to have a simple organisation. It is now known that cytoplasm has a highly complex organisation consisting of cytoplasmic matrix and several organelles. The motile cells have additional appendages for locomotion. The plant cells have a defined, rigid cell wall, which animal cells do not posseses; therefore, animal cells generally have an irregular shape.

Cell Wall

The plant cells have a fixed shape and size due to the presence of a cell wall, a specialised form of extra-cellular matrix, which is closely associated with the external surface of plant cell membrane. It is sufficiently thick, strong, and rigid and measures 0.1 nm to several micrometers. The composition and appearance of cell wall differs according to cell type and the function it performs. The fungal cell walls have a thick layer of polysaccharide fibres composed of either chitin or cellulose and a thin outer layer of mixed glycans. In the cell walls of algae, the

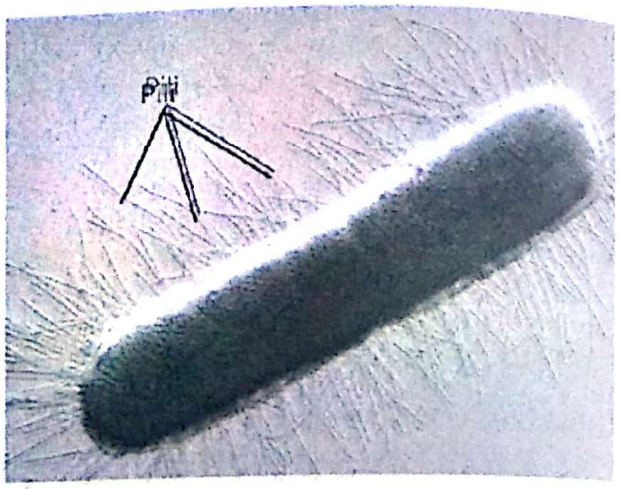

Fig. 9.6 *Pili-bacterial surface appendages*

substances commonly found are cellulose, galactans, mannans and minerals, such as silicon dioxide and calcium carbonate. In higher plants, the cell wall fibres are made up of the polysaccharide cellulose and are embedded in a highly cross-linked matrix of polysaccharides such as pectin, lignin and hemicellulose. Such a layer of pectin present in between the two cells is known as the **middle lamella** (Fig. 9.7a)

Primary Cell Wall

The primary wall in plant cells is composed of an intricate network of microfibrils in a gel-like matrix arranged in the following manner:

(i) ***Cellulose microfibrils***: The microfibrils are connected with the help of xyloglucan chains through hydrogen bonds. Thus a complete and continuous lattice is formed, which is embedded in a second network.

(ii) ***Pectic polysaccharides***: This constitutes second network, which is rich in galacto-uronic acid residue and forms cross links based on calcium bridges and other ionic interactions.

(iii) ***Structural proteins***: This is the third interlocking network, which consists of structural proteins that interweaves through the other two domains of network and forms wart and weft structure.

When a cell is young and small, the cellulose fibres are loosely packed and crosslinking of the cellulose fibrils is not complete. However, in mature cells due to crosslinking of fibres present in the wall, the cell is able to grow further. However, a new class of proteins called **expansin** has been found to be responsible for wall

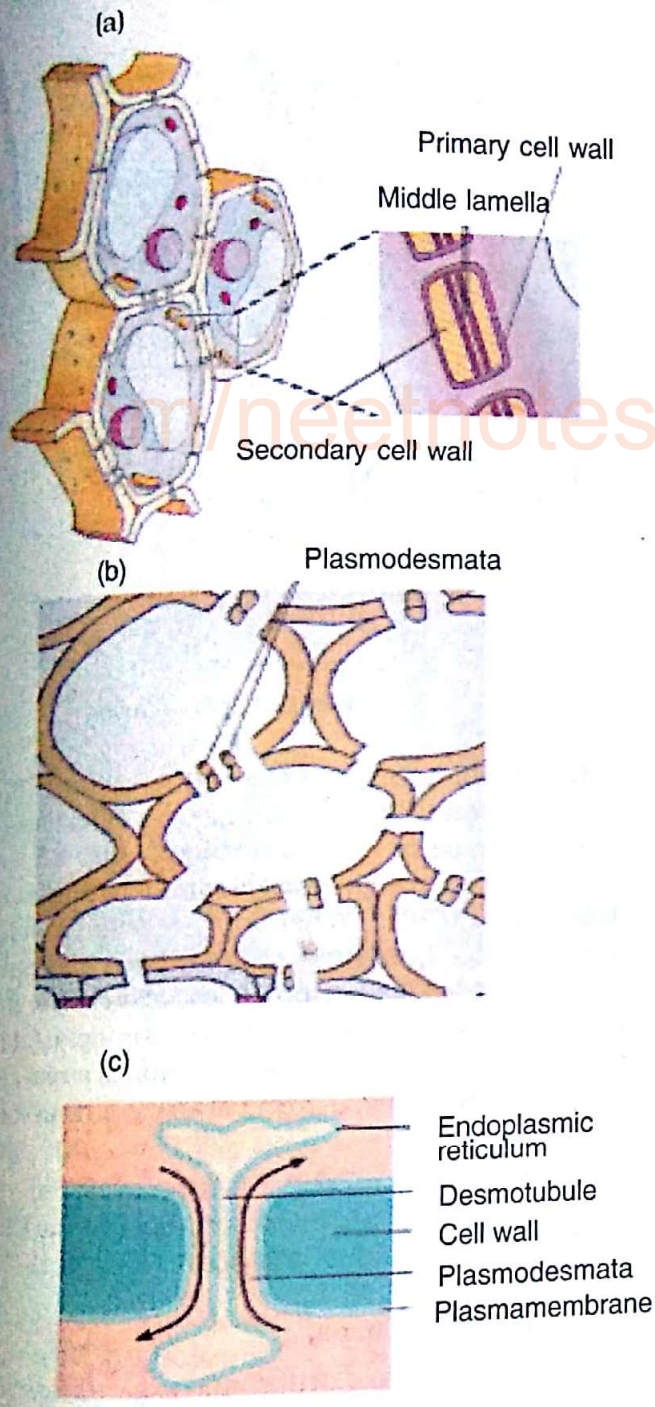

Fig. 9.7 *Cell wall showing (a) middle lamella primary cell wall and secondary cell wall (b) plasmodesmata (c) desmotubules*

loosening and cell expansion by addition of cellulose molecules to cellulose microfibrils.

Secondary Cell Wall

Inner to the primary wall the secondary wall is present, it is made up of cellulose fibrils in parallel rows at an angle with the other fibre bundles. Sometimes, hemicellulose, pectin and lignin may also be deposited in the secondary wall. The thick walls which have lignin deposits may also have pits, which are unthickened areas present in otherwise thick walls. Generally, the pits are present in pairs (on both the sides of wall). Therefore, these are called as pit pairs.

Tertiary Cell Wall

The cytoplasmic bridges present between adjacent cells are called **plasmodesmata** (Fig. 9.7b). It is a fine cytoplasmic canal, which is lined by plasma membrane and often has ER (endoplasmic reticulum) tubule called **desmotubule** (Fig. 9.7c). The plasmodesmata, help in maintaining continuity of living matter and the cytoplasm in such condition is often called **symplasm**. In contrast to this, the intercellular space containing non-living matter constituents is called **apoplasm**.

Plasma Membrane

It is a typical bilayer of lipids in which protein molecules are embedded and may also contain sterols of various kinds. Sterol molecules are more rigid than the phospholipids and, therefore, presence of sterols confers stability on eukaryotic membranes. These membranes are selectively permeable and so they work as barriers for certain molecules while allowing the entry of others.

Cytoskeleton

The ability of eukaryotic cells to adopt a variety of shapes and to carry out directed movements depends on the cytoskeleton. Three principal types of protein filaments: **microfilaments**, **microtubules**, and **intermediate filaments**, constitute the cytoskeleton. The microfilaments (Fig. 9.8) are ~8 nm in diameter, either scattered or organised into network or parallel arrays within the matrix. They are known to play a major role in cell motion or changes in shape. Such cellular movements associated with the microfibrils are movements of pigment granules, amoeboid movements and protoplasmic streaming. These microfilaments are made up of actin-like protein.

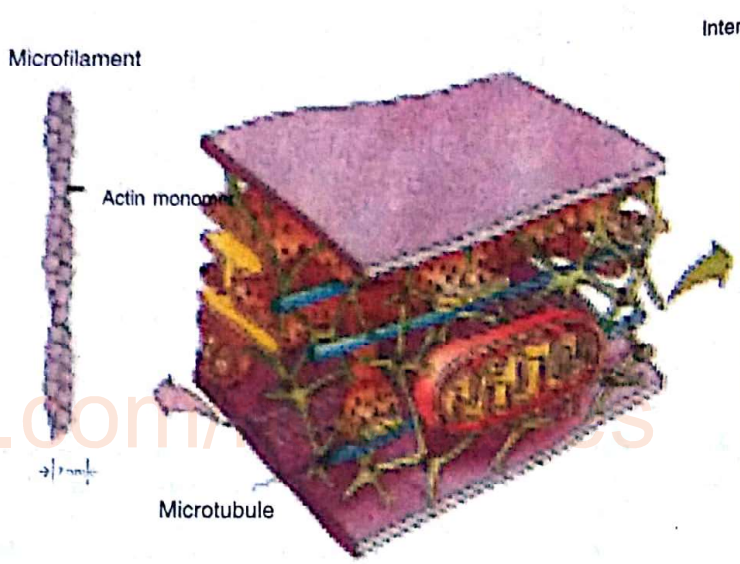

Fig. 9.8 *Microfilaments*

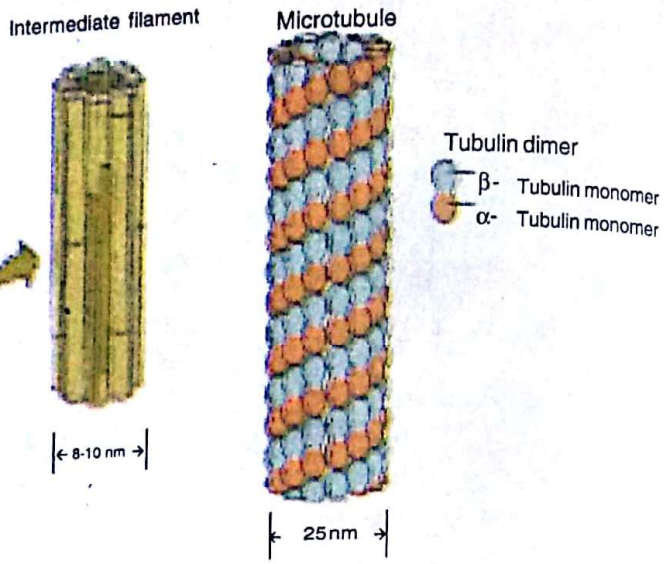

Fig. 9.9 *Microtubules*

The microtubules are hollow tubes of ~25 nm diameter formed from globular tubulin molecules (Fig. 9.9). The two subunits, alpha and beta, are arranged in a helical manner to form a cylinder. About 13 subunits are present in one turn or circumference. The microtubules perform following functions:

(i) help in maintaining cell shape,
(ii) are involved along with microfilaments in cell movements, and
(iii) participate in intracellular transport.

They play an important role in movement of organelles, e.g., movement of chromosomes. In such cells where microtubules are disrupted with chemicals like colchicines, no movement of chromosomes occurs. The intermediate filaments are tough and durable protein fibres or filaments of diameter between 8-10 nm, present in cytoplasmic matrix. In most animal cells, they form a basket around the nucleus and are present in cell-cell junctions.

Endoplasmic Reticulum

The term endoplasmic reticulum was used by Keith Porter (1953) to identify a fine reticulum (network) in endoplasm of the cell. It is an irregular network of membranous tubules, forming a continuous sheet enclosing a single internal space. In the cells that are involved in active secretion, the endoplasmic reticulum (ER) is studded on its outer surface with ribosomes and, because of its appearance such ER is referred to as **rough endoplasmic reticulum (RER)** (Fig. 9.10). Whereas the cells, which produce large quantities of lipid, have ER that is not studded with ribosomes. Such ER is known as **smooth ER (SER)**. The endoplasmic reticulum, apart from giving mechanical support to cytoplasmic matrix, also performs a number of functions:

(i) synthesis of secretory (serum proteins), lysosomal or membrane proteins on the ER membrane and its transport through ER lumen,
(ii) synthesis of lipids,
(iii) detoxification of drugs, and
(iv) associated with muscle contraction by release and uptake of Ca^{2+} ions.

Golgi Apparatus

It is a membranous organelle composed of flattened sac-like cisternae stacked on one another (Fig. 9.11). These cisternae resemble smooth ER. There are usually 4-8 cisternae in a stack. On the outer edges of the cisternae, a complex network of tubules and vesicles exists;

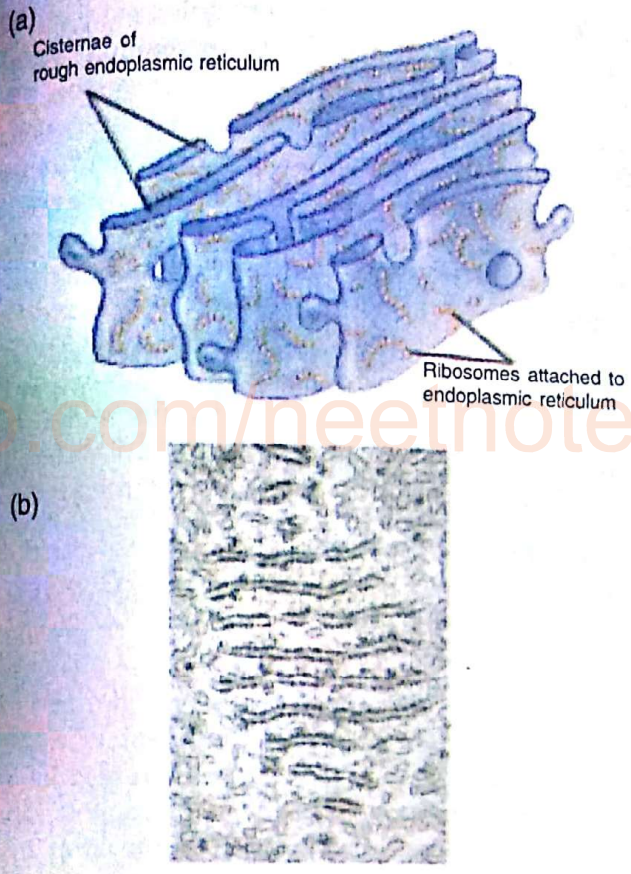

Fig. 9.10 *(a) Endoplasmic reticulum (b) Electron microscopic view*

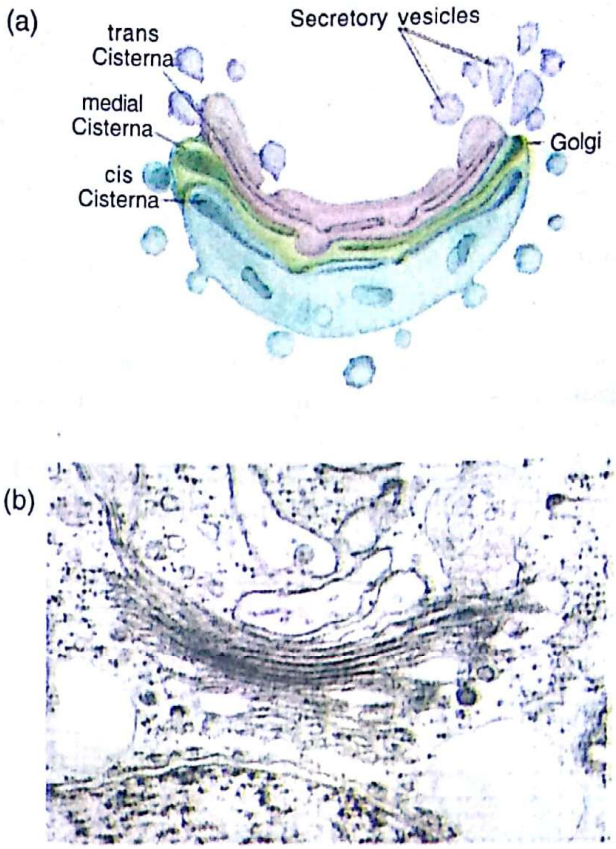

Fig. 9.11 *(a) Golgi apparatus (b) Electron microscopic view*

this structure appears to have a definite polarity. The two ends or faces are quite different from one another. The sacs on the forming face (*cis – face*) are associated with ER and differ from the sacs present on maturing face (*trans - face*) in thickness, its contents and the degree of vesicle formation. It appears that materials are transported from *cis* to *trans* face by vesicles that keep budding off from the cisternal edge to the next sac and so on. While most of the eukaryotic cells possess Golgi apparatus, many fungi and ciliated protozoans lack well-formed Golgi bodies. The cisternae in these organisms may have one stack or many. These stacks are also called **dictyosomes.**

The important function of Golgi apparatus is to package the material and prepare for secretions. The material to be secreted moves from ER to the Golgi apparatus, during which vesicles are budded off from the ER. These vesicles travel to Golgi apparatus and fuse with *cis* – cisternae. Therefore, Golgi apparatus is closely associated with ER in structural as well as functional aspects. Most proteins that are synthesised at ER, are glycoproteins which, after being transported to Golgi apparatus, are modified therein. Depending upon the target site, and the different fates, specific groups are added to the proteins here and then sent to their proper location enclosed in the vesicles that bud off from Golgi apparaturs.

Lysosomes

These are formed by budding off vesicles from Golgi apparatus. They are bound by a single membrane measuring about 500 nm. They are involved in intracellular digestion and contain the enzymes needed to digest all types of macromolecules. The enzymes present in such vesicles belong to the class hydrolases, which are active in acidic conditions. The acidic condition is maintained by pumping protons into the interior of lysosomes. The digestive enzymes are synthesised on RER and packed into the lysosomes. The portion of SER near the Golgi apparatus may also bud off lysosomes.

Most of the cells exhibit the phenomenon of **endocytosis** i.e. the import of materials (Fig. 9.12). This is of two major types, *viz.*, **phagocytosis** and **pinocytosis**. During phagocytosis large particles and even micro-organisms are ingested into a phagocytic vacuole or **phagosome**. In pinocytosis small amounts of surrounding liquid with its solute molecules are pinched off as pinocytotic vesicle or **pinosomes**. Phagosomes and pinosomes are collectively known as **endosomes**. On the other hand, cells are also involved in export of materials by secretion and this process is called **exocytosis**. The newly formed phagosomes fuse with primary lysosomes which contain hydrolytic enzymes and form **secondary lysosomes**. When the material in secondary lysosomes has been digested, the vesicle together with the remaining undigested material left behind is known as **residual body**. This is a defensive mechanism (for digesting the invading bacteria) and also helps in nutrition. Sometimes the cell may also digest a part of its own cytoplasm in a type of secondary lysosomes called **autophagic lysosomes**. The lysosomes accomplish this feat without releasing their digestive enzymes into the cell. The intricate complex of membranous organelles consisting of lysosomes, endosomes and associated structure alongwith Golgi apparatus seems to operate in a co-ordinated manner whose main function is the import and export of materials.

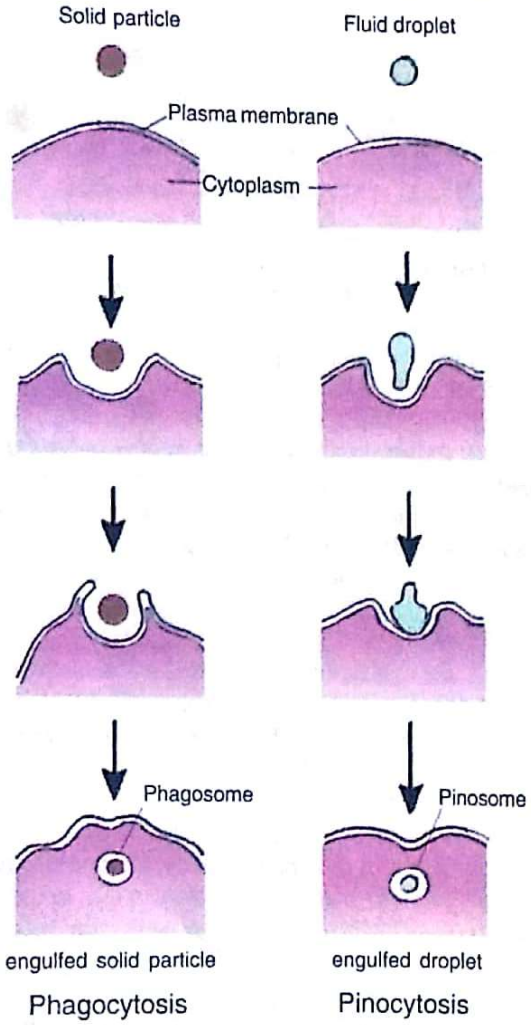

Fig. 9.12 Endocytosis

Cytoplasmic Vacuoles

These are non-cytoplasmic areas present inside the cytoplasm. They are supposed to be greatly expanded endoplasmic reticulum. The vacuoles of plant cells are bound by a single, semi-permeable membrane called **tonoplast**, whereas the vacuoles of animal cells are bounded by a lipoproteinaceous membrane. The vacuole can be classified into four types depending upon the contents and function they perform.

(i) **Sap vacuoles** (store and concentrate mineral salts as well as nutrients).
(ii) **Contractile vacuoles** (take part in osmoregulation and excretion).
(iii) **Food vacuoles** (contain digestive enzymes with the help of which nutrients are digested),and
(iv) **Air vacuoles** (present only in prokaryotes). They not only store metabolic gases but also help in buoyancy of cells.

Sphaerosomes

Sphaerosomes are single membrane bound spherical structures present in cytoplasmic matrix. They are associated with synthesis and storage of lipids. They develop from endoplasmic reticulum. In certain plants, the sphaerosomes may also posses lysosomal activity.

Microbodies

Microbodies are also single membrane bound organelles, associated with oxidation reaction other than those of respiration. These organelles often posseses a crystalloid core and granular matrix. There are two types of microbodies namely **peroxisomes** and **glyoxysomes**. The

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peroxisomes have enzymes for peroxide biosynthesis. These occur in most of the animals and plants, but are quite common in photosynthetic cells. Their number can be 70-100 per mesophyll cell wherein they interact with mitochondria and chloroplasts to take part in photorespiration. The glyoxysomes usually occur in fat-rich plant cells and are associated with triglyceride metabolism through glyoxylate cycle.

Ribosomes

The eukaryotic ribosomes may be associated with the endoplasmic reticulum or may be free in the cytoplasmic matrix. The ribosomes are of 80 S type consisting of 60 S and 40 S subunits. In general outline, the smaller subunit shows an obovate ellipsoid and cap-like structure, while the larger subunit appears dome-shaped. The smaller subunit has a platform, a cleft, a head and a base. The larger subunit, on the other hand, has a protuberance, a ridge and a stalk (Fig. 9.13).

When associated with ER, they are attached with their 60S subunit. The ER-bound ribosomes synthesise secretory proteins and lysosomal proteins, whereas the free ribosomes synthesize non-secretory proteins. Some proteins synthesised on ribosomes are transported to such organelles as nucleus, mitochondria and chloroplasts. The proper folding of proteins following synthesis is assisted by specific proteins called **chaperons**, which also assist in transport of proteins into organelles such as mitochondria. Several ribosomes may become attached to a single messenger RNA for simultaneous synthesis of variable protein molecules. Such complexes between messenger RNA and ribosomes are known as **polyribosomes** or **polysomes**.

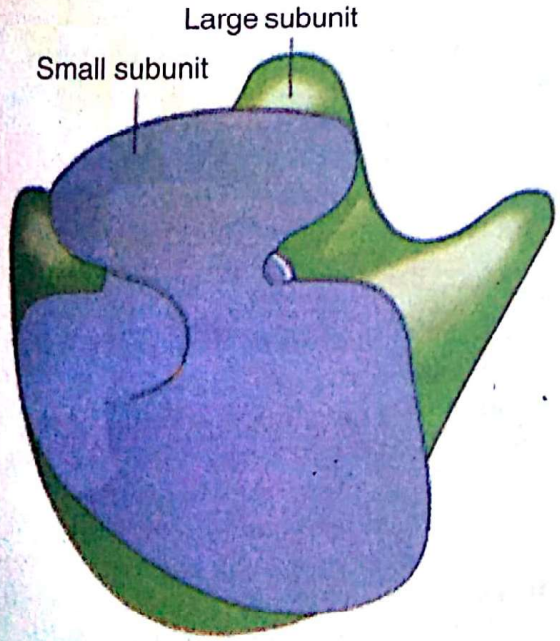

Fig. 9.13 *A ribosome*

Mitochondria

These are also called the powerhouse of the cell and are associated with generation of ATP (energy) through electron transport and oxidative phosphorylation. The mitochondria as (*sing.* mitochondrion) observed under electron microscope appear as cylindrical structures measuring approximately 0.6–2.0 nm in diameter and 5–10 nm in length. Usually the cells posses as many as 1000 or more mitochondria but some cells like those of yeast, and unicellular algae may have a single, giant tubular mitochondria twisted into a continuous network permeating the cell cytoplasm.

Mitochondria are bound by a double membrane, the outer membrane and the inner membrane, both separated by 6 – 8 nm inter-membrane space (Fig. 9.14). The inner membrane forms many folds called **cristae,** which increase its surface area. The shape of cristae is variable in mitochondria of different species, for example, fungal mitochondria often have plate-like cristae, while in *Euglena* cristae may be vesicle-shaped. The inner membrane encloses the mitochondrial matrix, which is quite dense and is loaded with ribosomes, DNA and often large calcium phosphate granules. Mitochondrial ribosomes are smaller than the cytoplasmic ribosomes, and are similar to the bacterial ribosomes in number of characters, e.g., size, composition of subunits, etc. Mitochondrial DNA is a closed circular molecule much like bacterial DNA. The mitochondrial matrix and the compartment is different from other organelles in chemical and enzymatic composition. The outer and inner membranes of mitochondria have different lipids. The enzymes and electron carriers for formation of ATP are located only in the inner membrane whereas the enzymes for TCA (tricarboxylic acid) cycle are located in the matrix. The inner membrane of mitochondrion has small spheres about 8.5 nm diameter, which are attached to

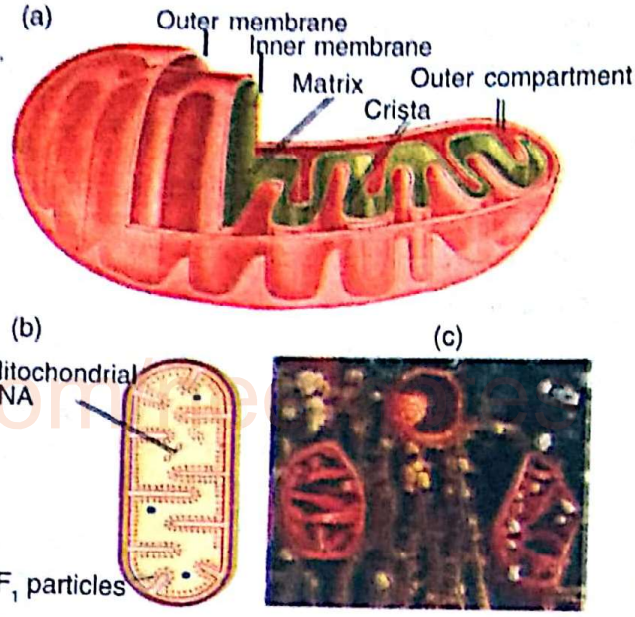

Fig. 9.14 *Structure of mitochondrion (a) Longitudinal section (b) Sectional view and (c) Electron microscopic view*

inner matrix of the membrane by stalks. These structures are called F_1 particles (elementary particles), and are associated with ATP production, during cellular respiration.

Presence of DNA and ribosomes in the mitochondria makes these organelles independent for production of some of their own proteins. However, most of the mitochondrial proteins are synthesised under the direction of cell nucleus. Multiplication of the mitochondria is by binary fission – again a prokaryotic, bacterial character. Another important feature of mitochondria is their character of changing structure. This is dependent upon the physiological activity of the cell as well as the activity occurring in the organelle. When ATP concentration is low or the respiratory chain is inhibited, the mitochondria are seen in an **inactive** or **orthodox state**. Under this condition, the matrix of the mitochondria occupies a larger area. In an **active** or the **condensed state** (when mitochondria are actively engaged in oxidative-phosphorylation and electron transport), the cristae are more randomly distributed and the inter-membrane space remains highly enlarged.

Plastids

The plastids are organelles that are found in only plant cells and some unicellular organisms (*Euglena*) of uncertain affinity. These can be classified into several types, depending upon the nature of pigments they contain. **Chloroplasts** contain chlorophyll pigments, whereas **chromoplasts** may contain pigments other than chlorophyll. The **leucoplasts** are devoid of any pigment, although they have the capacity to develop pigments also as and when required. These organelles are bound by two membranes. As these organelles contain their own genetic material, and protein synthesising machinery, i.e., DNA, RNA and ribosomes, they are capable of multiplication by a fission-like process.

Chlorophyll is the green coloured pigment present in the chloroplast. It performs the function of trapping light energy required for formation of two products, *viz.*, ATP and the reducing power molecules NADPH. Both these molecules are required for fixation of CO_2.

The chromoplasts synthesize and store other pigments like carotenoids, a group of pigments that include carotene and xanthophylls, which are responsible for yellow, orange or red coloration in plants. These also act as precursor of vitamin A in animal tissues.

The leucoplast (colourless plastids) act as storage organelles and are classified on the basis of the material stored, e.g., **amyloplasts** store carbohydrates in the form of starch, aleuroplasts (protein granules) store proteins, whereas the **elaioplasts** store oils or fats.

The chloroplasts are found in the leaves of green plants and are the most common and biologically important plastids. Each chloroplast is bound by a smooth outer membrane, which regulates the transport of material between interior of the organelles and the cytoplasm. The inner membrane runs parallel to the outer one and is extensively folded inward. The folding forms a series of parallel membranous sheets internally called **lamellae,** suspended in a fluid-like matrix called **stroma** (Fig.9.15).

The latter is made up of about 50 per cent soluble proteins, ribosomes, DNA and the machinery for protein synthesis. Most of the lamellae in the chloroplasts are organised to form sac-like structures, which have been termed as **thylakoids**. They are flattend vesicles arranged as a membranous network within the stroma. These thylakoids may be stacked like a pile of coins. This formation has been termed as

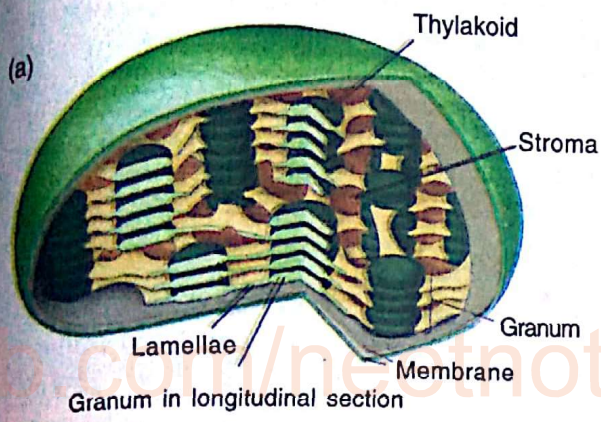

Fig. 9.15 *(a) Sectional view of Chloroplast (b) Electron microscopic view*

grana. In a typical chloroplast, as many as 40 – 60 grana may be present and each grana may have 2–100 small flattened thylakoids. It has been roughly estimated that more than 50 per cent of the chloroplast proteins and various components involved in the process of photosynthesis are present in thylakoids. In other words, the pigments, namely, chlorophyll, carotenoids and plastoquinone, are present in thylakoid membranes that are involved in photosynthesis.

Similarities Between Mitochondria and Chloroplasts

(i) It has been postulated that mitochondria and chloroplasts may have existed as independent organisms, which during evolution developed a symbiotic relationship with plant and animal cells and evolved into their present state.

(ii) Both mitochondria and chloroplasts originate and develop in the same way. They are formed by the division of pre-existing organelles.

(iii) Both are semi-autonomus organelles as both contain DNA, RNA and ribosomes. Both mitochondrial DNA (mtDNA) and chloroplast DNA (cpDNA) are circular in shape although cpDNA is much bigger than mtDNA. The genetic information contained in mtDNA and cpDNA is limited.

(iv) The symbiont hypothesis suggests that there are many similarities between prokaryotes on one hand, and mitochondria and chloroplasts on the other, e.g., the presence of circular DNA not associated with histones and 70 S ribosomes.

Nucleus

Nucleus is a relatively large organelle controlling all the activities of the eukaryotic cells (Fig. 9.16a). Some cells have more than one nucleus, e.g., **binucleate** cells have 2 nuclei per cell (*Paramoecium*), while **multinucleate** ones have many nuclei (*Ascaris*). However, some cells lack nucleus (**anucleate**) at maturity, such as, **mammalian RBCs** and **sieve tube cells** (food conducting elements in vascular plants). That the nucleus is a store house of hereditary information was proved by a Danish biologist Joachim Hammerling (1953) on the basis of his studies on *Acetabularia*.

EM studies have shown that nucleus is enclosed by two membranes, which form a **nuclear envelope**. The outer and inner membranes are separated by a narrow space called the **perinuclear space**. The outer membrane remains in continuation with endoplasmic reticulum and the inner one surrounds the nuclear contents. At certain places, the nuclear envelope is interrupted by the presence of small structures called pores (Fig. 9.16b). The pores are enclosed by circular structures called **annuli**. The pores and annuli together form the **pore complex**. The pores help in exchange of materials between **nucleoplasm** (nuclear fluid) and cytoplasm. RNA and ribosomes leave the nucleus through these pores. The nuclear envelope disappears during cell division and it reappears during nuclear reorganisation.

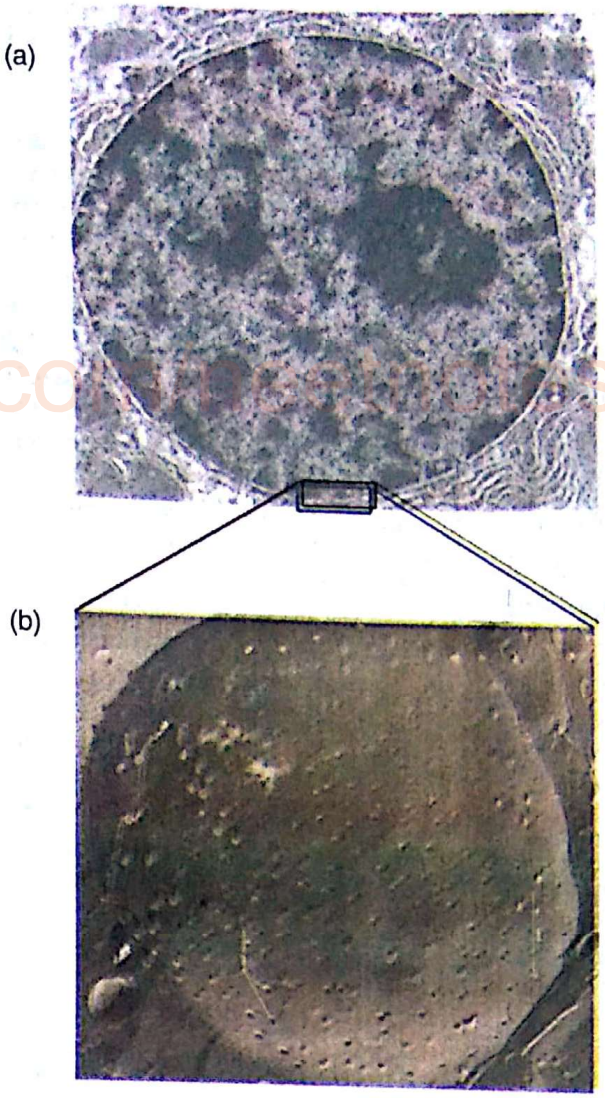

Fig. 9.16 *Structure of nucleus and nuclear envelope (a) Structure of nucleus (b) Nuclear envelope with pores as seen in electron microscope*

Nucleoplasm contains **nucleolus,** and **chromatin**. Nucleolus is a spherical structure, which is not separated from the rest of the nucleoplasm by a membrane. It is produced from and is associated with a specific nucleolar organising region of certain chromosomes. The nucleolus is the site for ribosomal RNA synthesis. The nucleoli are larger and more numerous in cells that are actively involved in protein synthesis.

During the interphase stage of the cell, the chromosomes are uncoiled in a loose, indistinct network called the **chromatin** that contains DNA, RNA and protein. The types of protein present and associated with DNA are histone and non-histone proteins. The chromosomes are thread-like structures, which become visible (under a light microscope) during cell division. In higher organisms, the well organised nucleus contains a definite number of chromosomes of definite size and shape. The chromosome shape (Fig. 9.17) is usually observable at metaphase and anaphase, when the position of primary constriction or centromere is clearly visible.

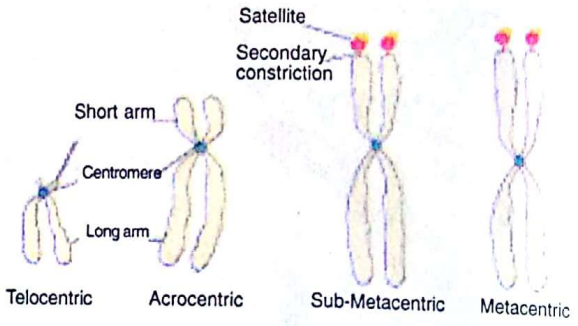

Fig. 9.17 *Types of chromosomes based on the position of centromere*

Based on the position of centromere, chromosomes are called:

(i) **Telocentric** – with terminal centromere,
(ii) **Acrocentric** – centromere is capped by a telomere,
(iii) **Submetacentric** – when centromere is subterminal in position, and
(iv) **Metacentric** – having median centromere.

Besides centromere, secondary constriction can also be observed in some chromosomes, which if present in the distal region of an arm, would pinch off a small fragment called **satellite**. The satellite remains attached to rest of the body of chromosomes by a thread of chromatin. The secondary constrictions are always constant in their positions and hence can be used as **markers**. The chromosomes having a satellite are marker chromosomes and are called **SAT chromosomes**.

Detailed study of chromosome morphology using light microscope revealed a coiled filament throughout the length of a chromosome. This is

called **chromonema**, that forms the gene-bearing portion of the chromosomes. It is a structure of a subchromatid nature. Electron microscope studies have revealed each chromatid to consist of a single chromatin fibre of ~30 nm diameter. In turn, each chromatin fibre contains a single DNA double helix.

Karyotype

All the members of a species of a plant or the animal are characterised by a set of chromosomes, which have certain constant characteristics. These may include the number of chromosomes, relative size, position of centromere, length of the arm, secondary constriction and satellites, features by which a particular set of chromosome can be arranged. This is called **karyotype** of a species. A diagrammatic representation of karyotype (morphological characteristic of the chromosomes) of a species is called **idiogram,** where the chromosomes of an organism are ordered in a series of decreasing size (Fig. 9.18).

Identificcation of chromosomes and karyotyping can be done by following techniques:

(i) **Banding technique-** the development of chromosome banding techniques have proved very useful for preparation of karyotypes in which linear differentiation due to distinct banding patterns allow identification of choromosomes that are similar in morphology. The banding technique allows detection of the regions containing repetitive DNA. A variety of different kinds of bands, e.g., **Q, C, G or R bands** have been used in **animal karyotyping.** Similarly, **C bands** and **N bands** have been used in **plant karyotyping**.

(ii) **Fluorescence *in situ* hybridisation (FISH)** and **multicolour fluorescence *in situ* hybridization (McFISH)** have been extensively used in animals and plants. Here using DNA probes, labelled with radioactive or nonradioactive molecules are used to locate the positions of specific DNA sequences on chromosomes. In McFISH, DNA may also be labelled with fluorochromes to allow the use of one or more colours to locate the position of one or

Metaphase plate

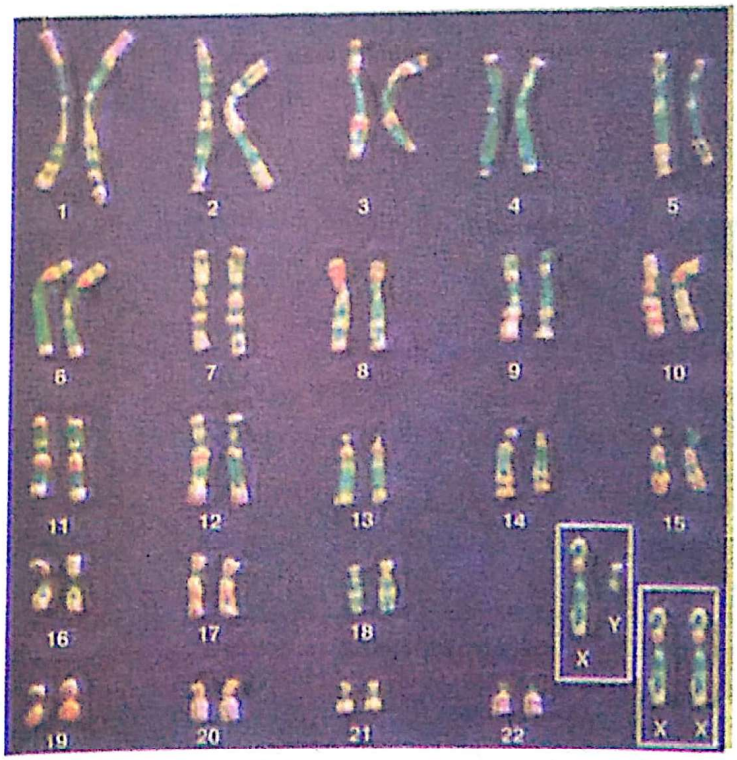

Fig. 9.18 *Karyotype of human chromosomes*

more DNA sequences simultaneously on the same chromosomes.

(iii) **Flow cytometry**: This is a recent technique, where a suspension of many thousands of chromosomes is made and the suspended chromosomes are stained with a DNA binding fluorochrome. As these chromosomes pass through the cytometer, the fluorescence is measured for individual chromosomes and the result is represented in the form of **histogram.** Each peak in this histogram represents one chromosome or a group of chromosomes of same size. This technique is so accurate that it allows detection of differences as small as 1.5 – 4.0 Mbp (mega base pairs). It helps in detection of **aneuploidy, duplication or deletion.**

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Uses of Karyotyping

(i) It suggests primitive or advanced features of an organism. If a karyotype shows a large size difference between the smallest and the largest chromosome of the set and having fewer metacentric chromosomes is called **asymmetric karyotype**, which is a relatively advanced feature.

(ii) The karyotypes of different species are sometimes compared and similarities in them are presumed to represent evolutionary relationship.

(iii) The flow cytometry technique allows detection of difference as small as 1.5–4.0 Mbp so that any variation in individual chromosomes, such as, duplication and deletion, can be easily detected.

Special Types of Chromosomes

In certain organisms there are special tissues where these chromosomes take up a special structure.

(i) **Lampbrush chromosomes** occur at the diplotene stage of meiosis in the **primary oocyte** nuclei of all animal species. Lampbrush chromosomes (Fig. 9.19) are also found in **spermatocytes** of several species, **giant nucleus of** *Acetabularia* and even in plants. These chromosomes are characterised by a remarkable change in structure. The change in structure includes an enormous increase in length.

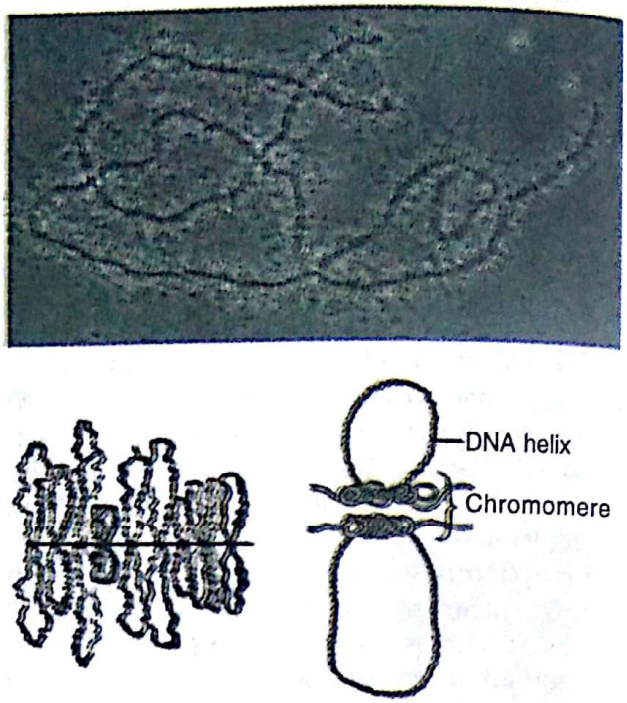

Fig. 9.19 *Structure of Lampbrush chromosome*

(ii) **Polytene chromosomes** occur in some cells of the larvae of the dipteran insects, such

Fig. 9.20 *Polytene chromosome*

as, *Drosophila*, mosquitoes and midges (*Chironomus*), which become very large, having high DNA content (Fig. 9.20). These cells are unable to undergo mitosis and are destined to die during metamorphosis. Polyteny of giant chromosomes is achieved by replication of the chromosomal DNA several times without nuclear division and the resulting daughter chromatids do not separate but remain aligned side by side.

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The Centriole

These are submicroscopic, microtubular, subcylindrical structures, which usually occur in the form of two granules. These are also called **diplosomes**. They are inside a specialised cytoplasm called **centrosphere** or **kinoplasm**. The complex formed of centriole and centrosphere was termed **centrosome** by Theodor Boveri (1888). The two centrioles lie

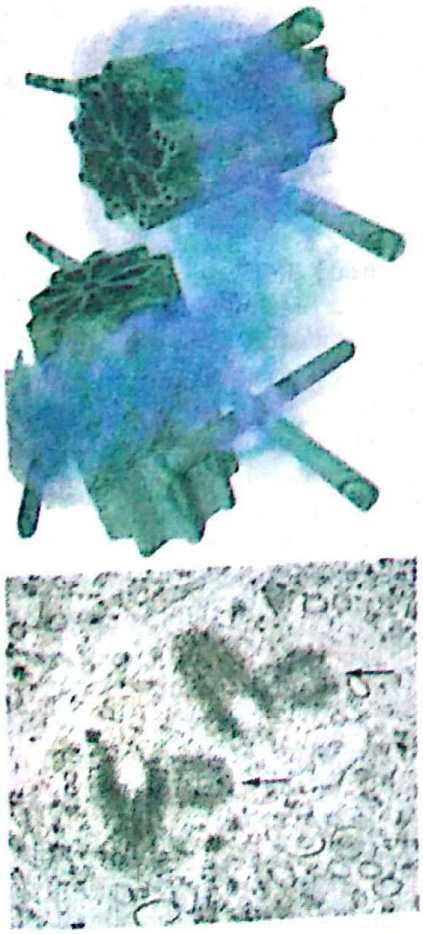
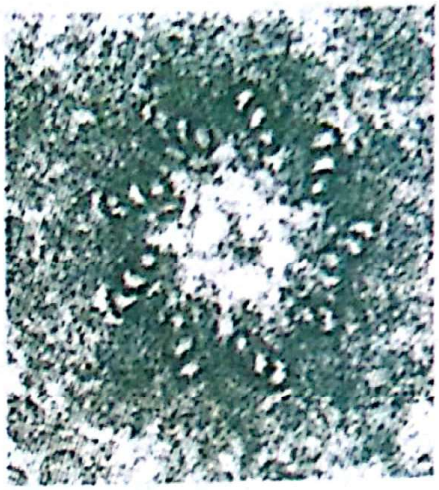


Fig. 9.21 *Centriole - Schematic (above) photograph (below)*



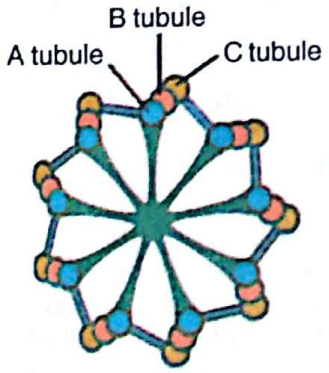


Fig. 9.22 *Centriole in transverse section*

at right angle to each other. Each centriole has a cartwheel organization having a whorl of nine peripheral triplet fibrils of tubulin (9 + 0) tilted at an angle of 40° (Fig. 9.21). The three subfibres of a triplet from outside to inside are designated as C, B and A (Fig. 9.22). Though each subfibre is expected to have 13 protofilaments like microtubules, both C and A subfibres share 2 – 3 protofilaments with B subfibres. Matrix fills the centrioles. The adjacent triplets are interconnected by proteinaceous linkers from C to A. The centre is occupied by a proteinaceous **hub**. The subfibre of each triplet is connected to the hub by means of radial proteinaceous strands called **spokes**. The spokes are also connected to C–A linkers with two types of thickenings X and Y.

The centrioles are surrounded by dense, amorphous, protoplasmic spheres. These help in formation of new centrioles in G_2 phase of cell

cycle. The centrioles are responsible for formation of basal bodies, cilia, flagella and astral spindle poles. These are mostly present in animal cells and flagellated organisms and structures (like spores, and gametes, etc.) but they are generally absent in plants.

Cilia and Flagella

The cilia and flagella are most prominent hair-like organelles on free surface of the cell and are associated with motility. Although both are whiplike and move the micro-organisms along, they differ from one another in two ways. Firstly, **cilia** are only 5-20 μm in length, whereas the flagella are 100 – 200 μm long. Secondly, their patterns of movement are usually distinctive. Flagella move in an undulating fashion and beat independently to generate planer or helical waves originating at either the base or the tip. If the wave moves from base to the tip, the cell is pushed along; a beat travelling from the tip towards the base pulls the cell through the water. Sometimes the flagellum will have lateral hair called flimmer filaments (thicker, stiffer hair) so that a wave moving down the filament towards the tip pulls the cell along instead of pushing it. Such a flagellum is referred to as a tinsel type flagellum, whereas the naked flagellum is referred to as whiplash flagellum. Cilia, on the other hand, normally have a beat with two distinctive phases. In the **power or effective stroke** (Fig. 9.23) the cilium propels through the surrounding fluids like an oar, thereby propelling the organisms along, in the water. The cilium next bends along its length, while it is pulled forward during the **recovery stroke** in preparation for another effective stroke. A ciliated microorganism actually coordinates the beat so that some of its cilia are in recovery phase, whereas others are carrying out their effective stroke. This coordination allows the organisms to move smoothly through the water.

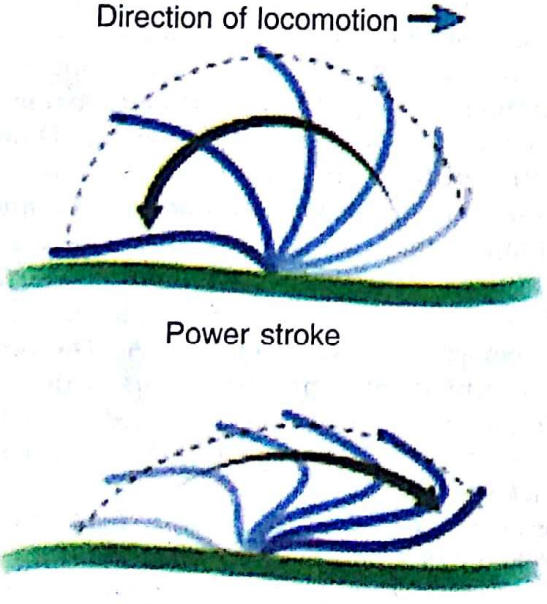


Fig. 9.23 *Movement of cilia*

Despite their differences, cilia and flagella are very similar in ultrastructure; both are membrane-bound cylinders. These organelles are a complex structure made up of four parts, *viz.*, shaft basal body, basal plate and rootlets, (Fig. 9.24). The **shaft** or the **axoneme**, consists of nine pairs of microtubule doublets arranged in a circle around two central tubules. This is called 9 + 2 pattern of microtubules. Each doublet also has pairs of arms projecting from subtubule A towards a neighboring doublet. A radial spoke extends from subtubule A towards the internal pair of microtubules with their central sheath. These microtubules are similar to those found in the cytoplasm. Each is made up of two types of tubulin subunits, the tubulins resemble the contractile protein actin in their composition.

A basal body lies in the cytoplasm at the base of each cilium or flagellum. It is short cylinder with nine microtubule triplets around its periphery (9 + 0 pattern) and is separated from the rest of the organelles by a basal plate. The basal body directs the construction of these organelles. Cilia and flagella appear to grow through the addition of preformed microtubule subunits at their tips.

Cilia and flagella bend because adjacent microtubule doublets slide along one another while maintaining their individual lengths. The doublet arms, about 15 nm long, are made up of the protein **dynein**. Adenosine triphosphate (ATP) powers the movement of cilia and flagella and isolated dynein hydrolyses ATP. It appears that dynein arms interact with the subtubule B of adjacent doublets to cause the sliding motion.

Cilia and flagella beat at a rate of about 10 – 40 strokes or waves per second and propel the microorganisms rapidly. Such speeds are equivalent to or are much faster than those seen in higher animals.

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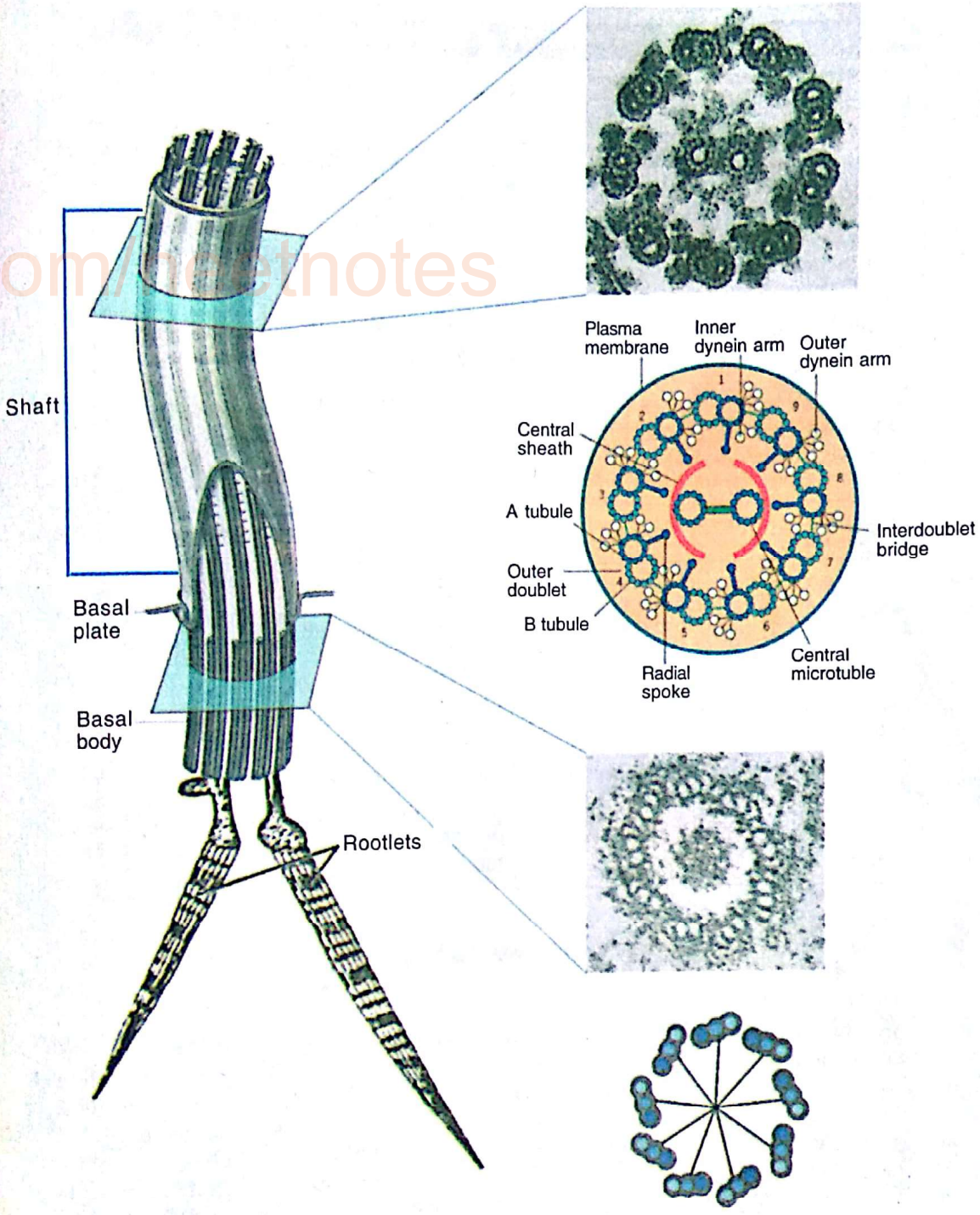

Fig. 9.24 *Section of cilia/flagella showing different parts*

The record holder is the flagellate *Monas stigmatica*, which swims at a rate of 260 µm/second (approximately 40 cell lengths per second). The ciliate protozoan *Paramoecium caudatum* swims at about 1500 µm or more per second (12 cell lengths per second).

The major differences between prokaryotic and eukaryotic cells are summarised in Table 9.2.

Table 9.2. Differences between Prokaryotic and Eukaryotic Cells

Prokaryotic cell	Eukaryotic cell
Lack of organised nucleus, the genetic material present in the form of nucleoid.	Nucleus well organised.
Nuclear membrane absent.	Nuclear membrane present.
DNA not complexed with histones.	DNA complexed with histones to constitute chromatin.
DNA in a circular form; not packed into chromosomes.	Linear DNA packed into well defined chromosomes.
Membrane-bound organelles absent.	Membrane-bound organelles like ER, golgi, lysosomes, etc. present.
Mitochondria absent.	Mitochondria present.
Chloroplast absent, photosynthetic lamellae may be present in photosynthetic bacteria.	Chloroplast present in plant cells.
Ribosomes only of 70S type.	Ribosomes of 80S (in cytoplasm) and 70S (in organelles) types in plant cells; and 80S and 55S types in animal cells
Cell wall made up of murein.	Cell wall absent in animal cells; in plant cells it is made up of cellulose, hemicellulose and lignin
Flagella simple, made up of flagellin and 9 + 2 organisation is absent.	Flagella specialised and show 9 + 2 organisation.
Cytoplasmic streaming not observed.	Cytoplasmic streaming is observed
Microtubules absent.	Microtubules-forming cytoskeleton present.
Different kinds of pili present.	Pili absent.

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SUMMARY

In general, two types of cells are recognised: prokaryotic and eukaryotic. Prokaryotic cells are much simpler in structure than the eukaryotic cells. Bacterium is an example of a prokaryote. In prokaryotes, the nuclear membrane, membrane-bound cell organelles, microtubules and cytoplasmic streaming are all absent. Their nucleus is simple: DNA is without histones and is packed into a nucleoid. The cell envelope of prokaryotes has three basic layers: (i) glycocalyx, (ii) the cell wall, and (iii) the plasma membrane. In order to explain the structure of cell membrane, the unit membrane concept was elaborated by Singer and Nicholson as fluid-mosaic model. The plasma membrane is the site for photosynthesis and respiration, synthesis of lipids and cell wall constituents in prokaryotes. As such it has a lot of enzymes and hormones required for these processes. The plasma membrane invaginates into the cell and forms vesicles of tubules or lamellae called mesosomes that provide larger surface area to plasma membrane for greater metabolic activity. Cytoplasm contains cell inclusions like polyphosphate, cyanophycean granules, glycogen granules, carboxysomes, metachromatic granules, volutin granules, sulphur granules, gas vacuoles.

ribosomes and nucleoid. Nucleoid is composed of a single circular molecule of double-stranded DNA, packed with nucleoid proteins. A flagellum is composed of three parts – filament, basal body and a hook. The fimbriae are small bristle like fibres, which attach bacteria to solid surface. The pili are elongated tubular structures, made up of a special protein pilin, which helps in the mating process.

The Eukaryotic cells have two major compartments nucleus and cytoplasm. The cell is enclosed by a cell wall, which is made up of cellulose and other substances like chitin (in fungus), pectin, mannans and minerals (in algae) and pectin and/or lignin, and hemicellulose (in higher plants). The primary wall is composed of cellulose microfibrils connected with xyloglucan chains in which pectin polysaccharide network is embedded and the structural proteins for third interlocking network. When the cell is undifferentiated, the fibres are loosely packed and cross-linking is not complete. However, as it matures, the cross-links are strengthened and the wall becomes rigid. The secondary wall is laid down on primary wall, and is made of cellulose, hemicellulose, pectin and lignin. In some plants another, the tertiary cell wall is laid down. The cell wall possesses minute cytoplasmic connection between adjacent cell called plasmodesmata. The organization of cytoplasmic membrane of eukaryotes is comparable to that of the prokaryotes.

The cytoplasmic matrix shows cytoplasmic streaming and contains a large amount of water and proteins. Micro-, macro- and intermediate protein filaments constitute the cytoskeleton. The latter maintains the cell shape, and is associated with cellular movements and intracellular transport.

Endoplasmic reticulum contains tubules or cisternae. They are of two types: rough and smooth. ER helps in the transport of substances, synthesis of proteins, lipoproteins and glycogens. The Golgi body is a membranous organelle composed of flattened sacs. The secretions of cells are packed in them and transported from the cell. Lysosomes are single membrane structures containing enzymes for digestion of all types of macromolecules. They are also called suicidal bags of cells. Sphaerosomes synthesise and store fats. Microbodies like peroxisomes are common in photosynthetic cells whereas the glyoxisomes are involved in fat metabolism.

Ribosomes are involved in protein synthesis. These occur freely in the cytoplasm or are associated with ER. They are made of two sub-units. Ribosomes associate with mRNA to form polyribosomes during protein synthesis.

Mitochondria help in oxidative phosphorylation and generation of adenosine triphosphate. They are bound by double membrane; the outer membrane is smooth and inner one folds into several cristae. It has its own DNA and ribosomes and is a semi-autonomous organelle. Plastids are pigment containing organelles found in plant cells only. They are two-membrane structures (outer smooth and inner extensively folded into thylakoids) and contain grana, which is suspended in a matrix called stroma. The grana is the site of light reaction and stroma of dark reaction. The green coloured plastids are chloroplast, which contain chlorophyll, whereas the other coloured plastids are chromoplasts, which may contain pigments like carotene and xanthophyll. Leucoplasts are colourless plastids. They are of three main types - amyloplast, aleuroplast and elaioplasts.

The nucleus is enclosed by nuclear envelope, a double membrane structure with nuclear pores. The inner membrane encloses the nucleoplasm and the chromatin material. The nucleolus is a spherical structure located in nucleus, which helps in RNA synthesis and ribosome subunit synthesis. Centrioles occur in animal cells in pairs, one being located at right angles to the other. It has a cartwheel like organisation with nine peripheral triplets of tubulin. The center is occupied by a protein rod hub and is responsible for formation of basal bodies, cilia, flagella and spindle fibres.

Cilia and flagella are associated with motility of cells. Flagella are longer than cilia. Flagella move by undulatory movement, whereas the cilia beat and propel. Flagella may be of tinsel or whiplash type. Both cilia and flagella are made up of four parts - the basal body, rootlets, basal plate and shaft. The pattern of microtubules is 9+2.

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CHAPTER 10

MOLECULES OF THE CELL

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By now, you have become familiar with the structure of the cell. You know that each component of the cell is responsible for a specific function. The cell components are made up of some basic molecules which help in complex interactions. The cell contains several molecules which perform a variety of functions. This collection of molecules is called **cellular pool**. The cellular pool consists of inorganic materials and organic compounds. Inorganic materials include salts, mineral ions and water. The main organic compounds are carbohydrates, lipids, amino acids, proteins, nucleotides, nucleic acids, hormones and vitamins. Some organic molecules remain in colloidal form in the aqueous intracellular fluid. Others exist in non-aqueous phases like the lipid membranes and cell walls. In animals, the cellular pool has a composition, which is quite different from that of the extracellular fluid. This distinction is maintained by the selective permeability of the plasma membrane. The cell maintains this pool by the intake and elimination of specific molecules. In this Chapter you will study about various materials and macromolecules of the cells.

10.1 INORGANIC SUBSTANCES

Salts, mineral ions and water are the important inorganic materials present in a cell. Let us study their occurrence and functions.

Mineral Elements

Most elements are combined as inorganic compounds in the Earth's crust as minerals. Mineral elements also occur in living organisms as components of organic and inorganic molecules and ions. In all body fluids there is a balance between the amounts of minerals present as ions and in complexes. Major minerals of the living organisms are calcium, phosphorus, sodium, chlorine, magnesium and sulphur. Some other minerals occur in very small amounts, e.g., iron, copper, cobalt, manganese, molybdenum, zinc, fluorine, iodine, and selenium.

The minerals essential for plant growth are divided into two groups. The mineral elements required in large amounts are termed **macronutrients** and include phosphorus, potassium, calcium, magnesium, sulphur and iron. Minerals required in trace amounts are known as **micronutrients**. These are manganese, cobalt, zinc, boron, copper and molybdenum. The same mineral can play a number of different roles.

Many mineral elements enter the structure of cellular components. Sulphur-containing amino acids also enter into proteins. Calcium imparts strength and rigidity to bones and teeth by getting deposited in them along with phosphates. So high are the contents of calcium and phosphorus in the bones, that bone dust is used as fertilizer. Calcium carbonate forms the exoskeleton of some invertebrates. Shells of some molluscs are burnt to yield lime (CaO). In plants, calcium pectate forms the middle lamella. Magnesium also contributes to the rigidity of bones and teeth. You are aware that iron deficiency leads to anaemia. This is because of a failure in haemoglobin synthesis. Iron (Fe^{2+}) combines with the pigment porphyrin to form *heme*. The latter binds with different proteins to form respiratory pigments that help in respiration. These include the oxygen-carrying pigment **haemoglobin** of erythrocytes and the oxygen-storing pigment **myoglobin** of muscle cells. **Cytochromes** are heme-protein complexes acting as oxidizing agents. The plant pigment chlorophyll is a complex of magnesium (Mg^{2+}) and porphyrin along with a long lipid tail. Iodine occurs in mammals as inorganic iodine, protein-bound iodine of blood and thyroid hormones.

Dietary deficiency of iodine depresses thyroid activity and enlarges the thyroid gland (**goitre**). The soils of northern hill regions of India are deficient in iodine; so are the crops and drinking-water. Consequently, goitre is widespread among the people of these regions. Steps are taken to overcome this disorder by supplying iodised common salt.

Mineral elements present in trace amounts are mostly required for enzyme action. For instance, manganese is required for the activity of enzymes needed for the synthesis of oligosaccharides and glycoproteins. Mitochondria are rich in manganese. Molybdenum is necessary for fixation of nitrogen catalysed by the enzyme nitrogenase. Copper occurs in cytochrome oxidase. Magnesium is essential for a large number of enzymes, particularly those utilising ATP.

Sodium and potassium are responsible for the maintenance of extracellular and intracellular fluids through the osmotic effects of the concentrations. These two ions are also responsible for the maintenance of membrane potential. Na^+ and K^+ ions are also responsible for the transmission of electrical impulses in the nerve cells. However, calcium and magnesium reduce the excitability of nerves and muscles. Both in cells and extracellular fluids diabasic phosphate (HPO_4^{2-}) and monobasic phosphate ($H_2PO_4^-$) act as acid-base buffers to maintain the H^+ ion concentration of cellular fluids.

Water

You might recall that 70 - 90 per cent of living cells is water. In humans, about two-third of the body is formed of water and, of these, about 55 per cent (20-22 litres) is confined to cells as intracellular water. The remainder is found in extracellular fluids like blood, tissue fluid and lymph.

Water is a tiny polar molecule and passes readily through the membranes. You can recollect how in a water molecule the oxygen and hydrogen atoms with opposite polarity are bonded by hydrogen bond (see Fig. 2.4b). Two electronegative atoms of oxygen of two water molecules share a hydrogen atom and become linked by hydrogen bond. In this manner, several water molecules are hydrogen bonded to form short-lived macromolecular aggregates. Such hydrogen bonding of several water molecules results in the formation of a lattice structure. Fluidity of water is maintained by very rapid formation and dissociation of hydrogen bonds between water molecules.

Water acts as a solvent in living organisms. To dissolve in water, a solute molecule must form hydrogen bonds with water molecules to join this lattice structure. Hydrophilic polar groups of solutes can form hydrogen bonds with water molecules. So, polar molecules join the lattice structure of water and dissolve in it. This solvent action of water enables the dissolved polar solutes such as glucose to diffuse uniformly in cytoplasm and to be transported in extra-cellular fluids. But hydrophobic non-polar groups can not join the lattice structure of water. So non-polar molecules such as fats do not dissolve in water and have to be transported in water in association with polar molecules.

Water maintains molecular conformations and stabilises structural organisation of living organisms. In aqueous body fluids, proteins and nucleic acids become folded and coiled to assume specific 3-dimensional forms suitable for biological activity. Interaction between water and phospholipid molecules arranges the latter into the lipid bilayer of the membrane. Water ionizes to a small but significant extent into H^+ and OH^-, depending on temperature. Proteins, nucleic acids and phospholipids attain specific ionic states by accepting or donating H^+ from or to water. Mineral salts also ionize in an aqueous medium of the cells. Owing to its solvent action, water forms an ideal medium for chemical reactions. The solute molecules and ions intermix and make intimate contact with each other. Water is a reagent and is a source of H^+ and OH^- for many biochemical reactions. During photosynthesis, water donates the electron to chlorophyll and is oxidised to molecular oxygen.

Water helps in maintaining the constancy of the internal environment of an organism. Some substances, capable of neutralising acids or bases, remain in solution in the cytoplasm as extracellular fluids e.g., bicarbonate (HCO_3), carbonic acid, dibasic phosphate (HPO_4^{2-}) and monobasic phosphate ($H_2PO_4^-$). Acids and bases mix in the body fluids with these substances and are neutralised by them. Due to solvent action water aids in maintaining a constant pH. The high specific heat and mobility of water

distribute heat uniformly throughout the body. Water also helps in heat loss from skin and plant surfaces. High latent heat of vapourisation of water causes elimination of excess heat through evaporation. This maintains a constant body temperature. Elimination of waste products through urine also helps in homeostasis.

10.2 ORGANIC COMPOUNDS

Organic or biological molecules may be small and simple. Often these simple molecules assemble and form large and complex molecules, called macromolecules. These include four main classes: **carbohydrates, lipids, proteins** and **nucleic acids**. All macromolecules except lipids are formed by the process of **polymerisation**, a process in which repeating subunits termed **monomers** are bound into chains of different lengths. These chains of monomers are called **polymers**. The large size and complex 3-dimensional shape of macromolecules enables them to function as structural components, molecular messengers, energy sources, enzymes, nutrient stores and sources of genetic information.

Carbohydrates

Carbohydrates represent combinations of carbon, hydrogen and oxygen. Although carbohydrates can be generally represented by the empirical formula $(CH_2O)_n$, some carbohydrates contain additional atoms of sulphur or nitrogen. In these molecules, the carbon forms chains or rings with two or more hydroxyl groups and either an aldehyde or a ketone group, giving them the technical definition of polyhydroxy aldehydes or ketones.

These molecules exist in a great variety of configurations. The common term **sugar** (saccharide) refers to a simple carbohydrate such as a **monosaccharide** or **disaccharide** that has a sweet taste. A monosaccharide is a simple polyhydroxy aldehyde or ketone molecule containing 3–7 carbons (trioses,tetroses,pentoses, etc.) A disaccharide (Fig.10.1) is a combination of two monosaccharide molecules, and a **polysaccharide** is a polymer of many monosaccharides bound in linear or branched chain patterns. Monosaccharides and disaccharides are specified by combining a prefix that describes some characteristics of the sugar with the suffix –ose. For example, hexoses are composed of 6 carbon, and pentoses contain 5 carbon. Glucose is the sugar form of fruits, and xylose a pentose sugar, derives its name from the Greek word (*xylon*) for wood. Disaccharides are named similarly; **maltose** means malt sugar **lactose** is an important sugar component of milk, and **sucrose** is the common sugar (cane sugar).

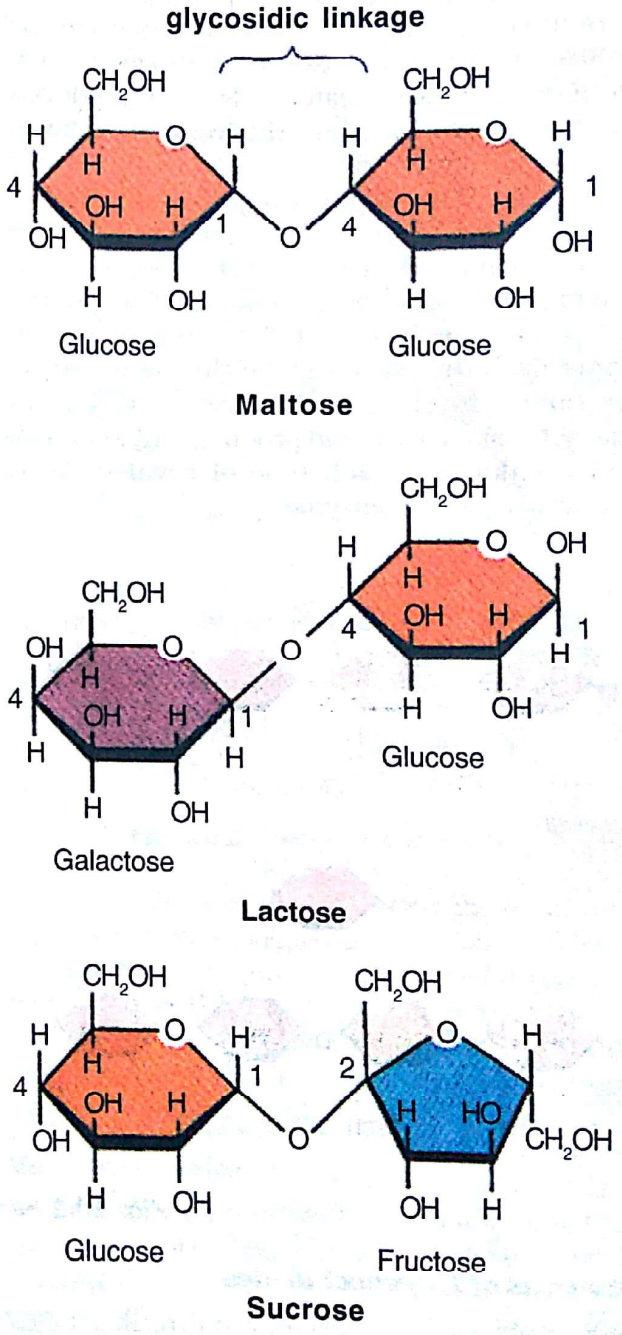

Fig. 10.1 Disaccharide molecules

Carbohydrate Bonds

The subunits of disaccharide and polysaccharide are linked by means of glycosidic bonds, in which carbon of one sugar unit is bound to carbon of another suger unit through the oxygen atom. Maltose is formed when the number one (1) carbon on a glucose bonds to the (4th) carbon on a second glucose. Lactose is formed when galactose and glucose join by their (1) and (4) carbons. Sucrose is formed when glucose and fructose join between their (1) and (2) carbons. Forming this bond, one carbon gives up its OH groups and the other loses the hydrogen from its OH groups. Since a water molecule is produced, this reaction is known as **dehydration synthesis**, a process common to most polymerisation reactions. Three polysaccharides (starch, cellulose and glycogen) are structurally as well as biochemically distinct, even though, all are polymers of the same monosaccharide glucose(Fig.10.2). The basis for their differences lies primarily in the exact way the glucose molecules are bound together, which greatly affects the characteristics of the end product. The synthesis and breakage of each type of covalent bond requires a specific enzyme.

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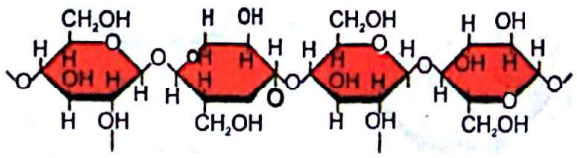
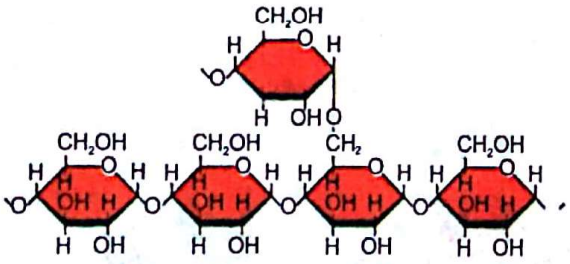
plants and many microscopic algae derives its strength and rigidity from cellulose. Because of this role, cellulose is the most abundant organic substance on Earth. Only a few bacteria, fungi and protozoa can digest it. Other structural polysaccharides can be conjugated to amino acids, nitrogen bases, lipids or proteins. **Agar**, a common polysaccharide used for preparing solid culture media, is a natural component of certain seaweeds. It is a complex polymer of galactose and sulphur-containing carbohydrates. The **exoskeleton** of insects and crustaceans and the cell wall of certain fungi contain chitin, a polymer of glucosamine (a sugar with an amino functional groups). **Peptidoglycan** is one special class of compounds in which polysaccharides (glycans) are linked to peptide fragments (a short chain of amino acids). This molecule provides the main source of structural support to the bacterial cell wall. The cell envelope of gram negative bacteria also contains lipopolysaccharide, a complex of lipids and polysaccharide responsible for inducing symptoms such as fever and shock.

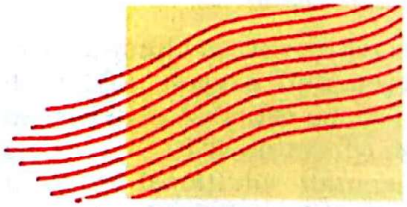
The outer surface of many cells has a delicate 'sugar coating' composed of polysaccharides



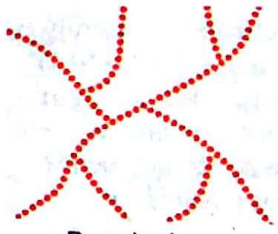
Cellulose



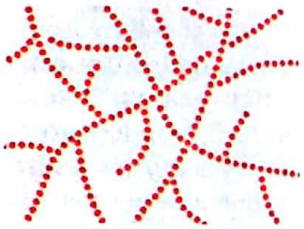
Starch and glycogen



Linear strands of cellulose molecules



Branched starch molecule



Highly branched glycogen molecule

Fig. 10.2 *Starch and glycogen*

Functions of Polysaccharides

Polysaccharides typically contribute to structural support and protection, and serve as nutrient and energy stores. The cell wall in bound to proteins in various ways. This combination is called mucoprotein or glycoprotein. This structure, called **glycocalyx** functions in attachment to other cells or as a

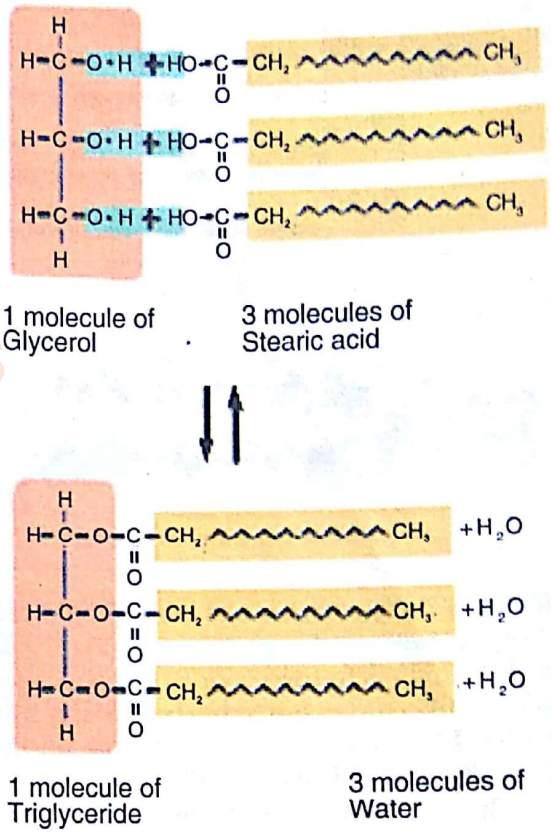
site for receptors – surface molecules that receive and respond to external stimuli. Small sugar molecules account for the differences in human blood types, and carbohydrates are components of large protein molecules called antibodies. Many bacteria are coated by a protective polysaccharide capsule that contributes to their infectiousness. Some viruses have glycoproteins on their surface with which they adhere to and invade their host cells. Polysaccharides are usually stored by cells in the form of glucose polymers such as **starch** or **glycogen**. Organisms with the appropriate digestive enzymes only can break them down and use them as a nutrient source. Since a water molecule is required for breaking the bond between two glucose molecules, this is also termed as **hydrolysis**. Starch is the primary storage food of green plants, microscopic algae and some fungi. Glycogen (animal starch) is a stored carbohydrate in animals and certain groups of bacteria and fungi.

Lipids

The term lipid is derived from the Greek word *lipos*, meaning fat. It is an operational term for a variety of substances that are not soluble in polar solvent such as water but dissolve in non-polar solvents such as benzene, ether and chloroform. This occurs because lipids contain relatively long hydrocarbon chains that are non-polar and thus hydrophobic. The main groups of compounds classified as lipids are **monoglycerides, diglycerides, triglycerides, phospholipids, steroids** and **waxes**.

Triglycerides

One important group of stored lipids is triglycerides, a category that includes **fats** and **oils**. Triglycerides are composed of a single molecule of glycerol bound to three fatty acids (Fig. 10.3). **Glycerol** is a 3 carbon alcohol with 3 OH groups that serve as binding sites. **Fatty acids** are long chain hydrocarbon molecules with a carboxyl group (COOH) at one end which binds to one of the OH groups of the glycerol, thus forming a bond called **ester bond**. The hydrocarbon portion of a fatty acid can vary in length form 4 – 24 carbons. The fat may be saturated or unsaturated. If the carbons in the chain are single bonded, the fat is saturated; if there is at least one C = C double bond in the



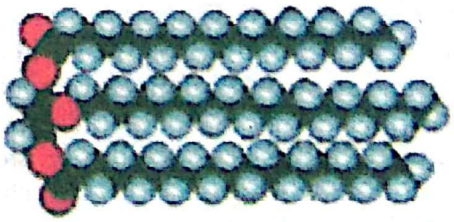


Fig. 10.3 Triglyceride molecule

chain, it is unsaturated. The structure of fatty acids is responsible for the physical nature of fats and oils (liquid fats) which are greasy and insoluble. In general, solid fats are saturated, and oils are unsaturated. In most cells for long term, triglycerides are stored in concentrated form as droplets or globules.

Membrane Lipids

A class of lipids that serve as major structural component of cell membranes is phospholipids. Although phospholipids are similar to triglycerides in containing glycerol and fatty acids, there are some significant differences. Phospholipids contain only two fatty acids

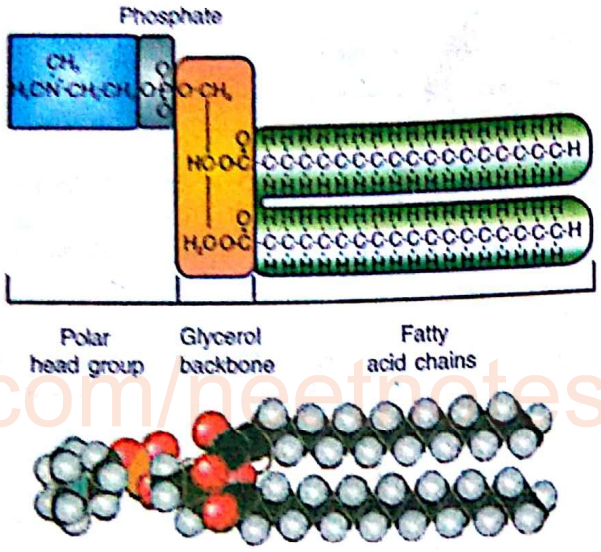


Fig. 10.4 *Phospholipid*

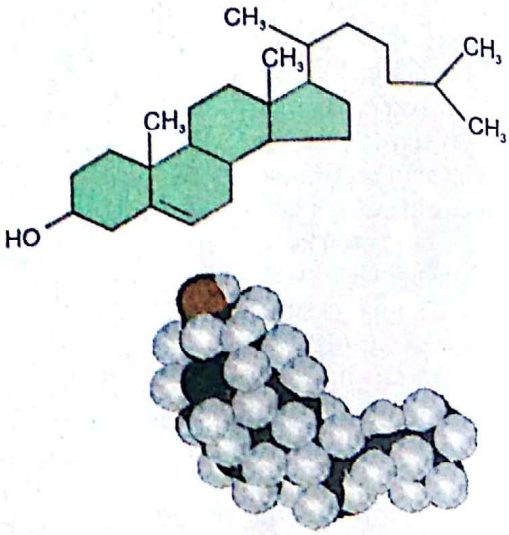


Fig. 10.5 *Cholesterol*

attached to the glycerol, while the third glycerol binding site holds a phosphate group (Fig. 10.4). This phosphate is in turn bonded to an alcohol. These lipids have both **hydrophilic** and **hydrophobic** regions due to a charge on the phosphoric acid / alcohol 'head' of the molecule and lack of a charge on the long 'tail' of the molecule (formed by the fatty acids). When exposed to an aqueous solution, the charged heads are attracted to the water phase and the non-polar tails are repelled from the water phase. The way lipids naturally assume single and double layered (bilayer) configuration makes them a valuable component of the primary framework of cell membranes. When two single layers of polar lipids come together to form a double layer, the outer hydrophilic face of each single layer will orient itself towards the solution and the hydrophobic portion will become immersed in the core of the bilayer. The structure of lipid bilayer helps the membrane in functions such as selective permeability and fluid nature.

Steroids

These are complex compounds commonly found in cell membranes and animal hormones. The best known of these is the sterol called **cholesterol** (Fig. 10.5) which reinforces the structure of the cell membrane in animal cells and in an unusual group of cell-wall deficient bacteria called **mycoplasmas**. The cell membrane of fungi also contains a sterol called **ergosterol**. **Prostaglandins** are fatty acid derivatives found in trace amounts that function in inflammatory and allergic reactions, blood clotting and smooth muscle contraction.

Wax

Waxes are esters formed between a long chain alcohol and saturated fatty acids (Fig. 10.6). This material is typically pliable and soft when warm but hard and water resistant when cold (e.g., paraffin). Fur, feathers, fruits, leaves, human skin and insect exoskeleton are naturally waterproofed with a coating of wax. Bacteria that cause tuberculosis and leprosy produce a wax (wax-D) that contributes to their pathogenicity.

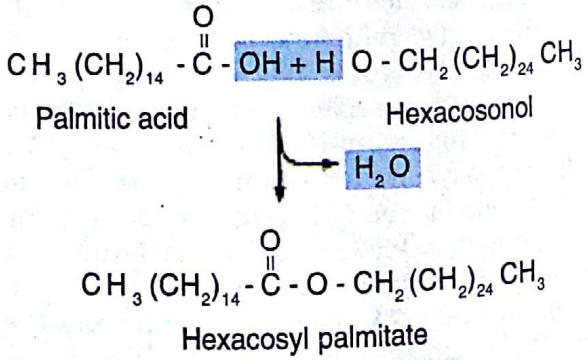


Fig. 10.6 *Beewax*

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Proteins

One of the predominant organic molecules in cells proteins, which are chemically and physically most diverse. The term protein was coined by Gerardus Johannes Mulder (1802–1880) and is derived from the Greek word *proteios* meaning 'of the first rank'. To a large extent the structure, behaviour and unique qualities of all living beings are the consequences of the protein they contain. The building blocks of proteins are **amino acids**, which exist in about 20 different naturally occurring forms. Various combinations of these amino acids account for nearly infinite variety of proteins. All amino acids have a basic skeleton consisting of a carbon (α carbon) linked to an amino group (NH_2), a carboxyl group (COOH), and a hydrogen atom (H). The variations among the amino acids occur at the R group, which is different in each amino acid. It imparts the unique characteristic to the molecule and to the protein that contains it. A covalent bond called **peptide bond** (Fig. 10.7) forms between the amino group of one amino acid and the carboxyl group of another amino acid. This type of bonding makes it possible to produce molecules varying in length from two amino acids to chains containing thousands of them. Peptide usually refers to a molecule composed of a short chain of amino acids, such as a dipeptide (two amino acids), a tripeptide (three) and a tetrapeptide (four). A **polypeptide** contains unspecified number of amino acids, but usually has more than 20. It is often a smaller subunit of a protein. A protein is the largest of this class of compounds and usually contains a minimum of 50 amino acids. The terms polypeptide and protein are often interchangeably used, though not all polypeptides are large enough to be considered proteins.

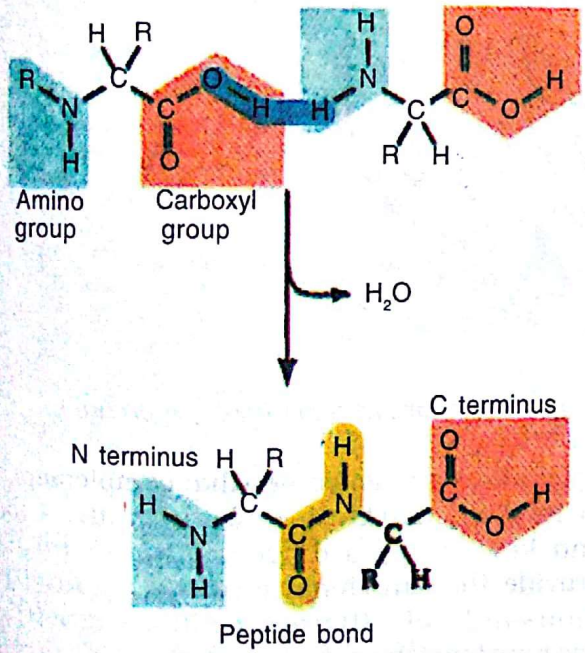

Fig. 10.7 *Peptide bond*

Structure of Proteins

Protein is synthesised on the ribosome as a linear sequence of amino acids which are held together by peptide bonds. Just after the synthesis is completed, the protein folds into a specific three dimensional form. According to the mode of folding, four levels of protein organisation (Fig. 10.8) have been recognised i.e., **primary, secondary, tertiary** and **quaternary**. Primary protein structure is correctly described as the type, number and order of amino acids in the peptide chain (Fig. 10.8a). The first protein to have its primary structure determined was insulin, the pancreatic hormone that regulates glucose metabolism in mammals. Secondary protein structure arises when various functional groups exposed on the outer surface of the molecule interact by forming hydrogen bonds. This causes the peptide to twist into a coiled configuration called the α-helix or many peptide chains to fold into a flat, β-pleated sheet (Fig. 10.8b). Some proteins contain both types of secondary configurations. Tertiary protein structure arises when the secondary level proteins undergo further folding by additional bonds between functional groups, such as disulfide bonds between sulfur atoms on two different cysteins of protein molecule (Fig. 10.8c). Quaternary protein structure may be assumed by some complex proteins in which more than one polypeptide forms a large multi unit protein (Fig. 10.8d). The quaternary structure in proteins that are composed of two or more polypeptide chains, refers to the specific orientation of these chains with respect to one another and the nature of interactions that stabilise this orientation.

The individual polypeptide chains of the protein are called subunits and the active protein itself

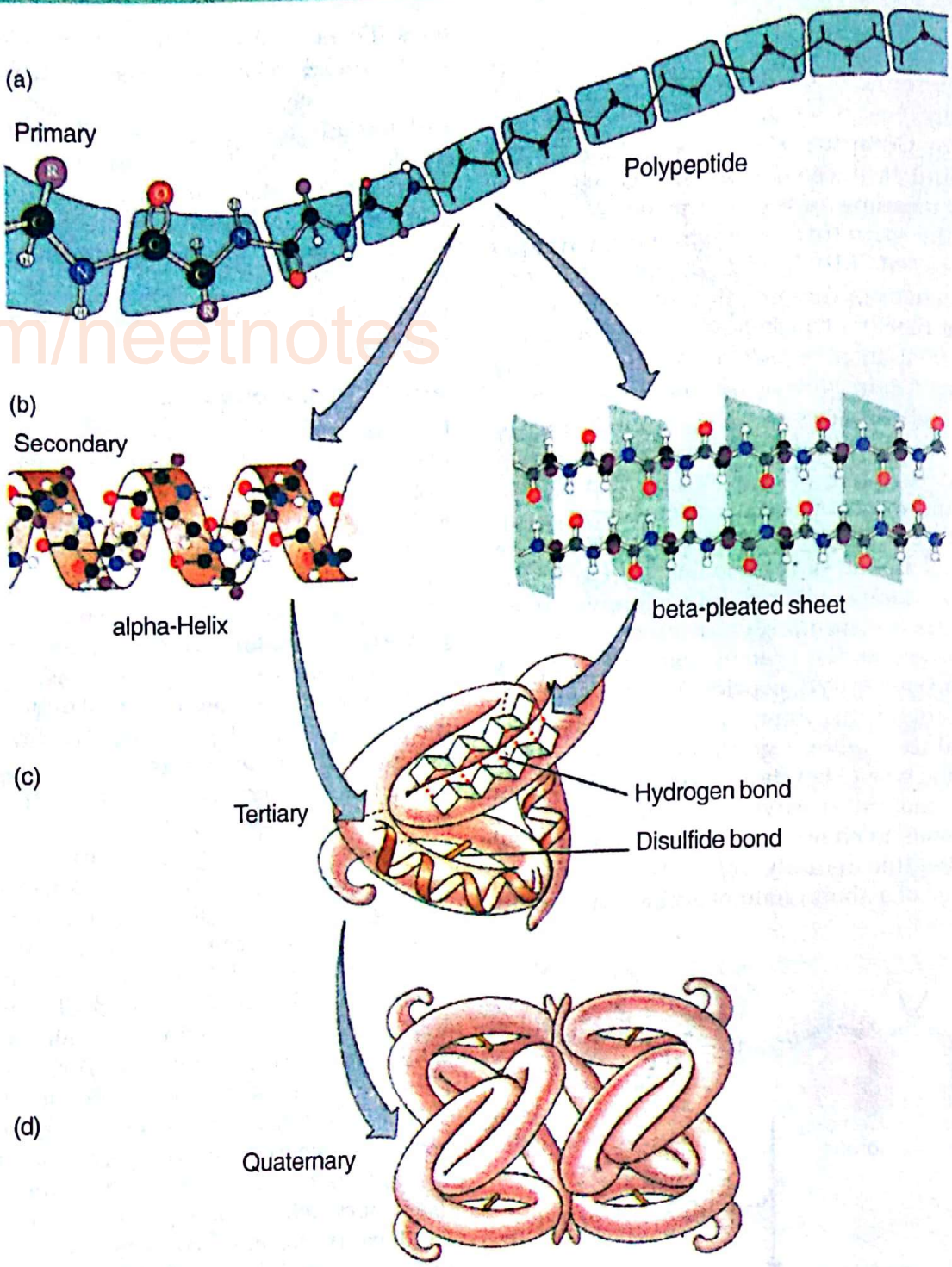

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Fig. 10.8 *Levels of protein organization (a) primary (b) secondary (c) tertiary (d) quaternary*

is called multimer. The multimeric proteins containing up to 32 subunits have been described. The most common multimers are dimers, trimers, tetramers, pentamers and decamers.

The most important outcome of intra-chain bonding and folding is that a protein can react only with molecules that complement or fit its particular surface features like a lock and key. Such a degree of specificity can provide the functional diversity required for thousands of different cellular activities. The functional three dimensional form of a protein is termed the **native state**, and if it is disrupted

by some means, the protein is said to be denatured. Agents such as soap, detergents, acid, alcohol and some disinfectants disrupt the stabilising inter-chain bonds and cause the molecule to become non-functional.

The Nucleic Acids

You are aware that there are two kinds of nucleic acids, namely, **deoxyribonucleic acid** (DNA) and **ribonucleic acid (RNA)** (see Chapter 2). These were originally isolated from the cell nucleus. Shortly thereafter they were also found in cells with no defined nuclei (bacteria), and in viruses. DNA contains a special, coded genetic programme with detailed and specific instructions for each organism's heredity. We briefly consider here, the structure and some functions of DNA, RNA, and a close relative ATP (adenosine triphosphate). Both nucleic acids are polymers of repeating units called **nucleotides**,(Fig.10.9),

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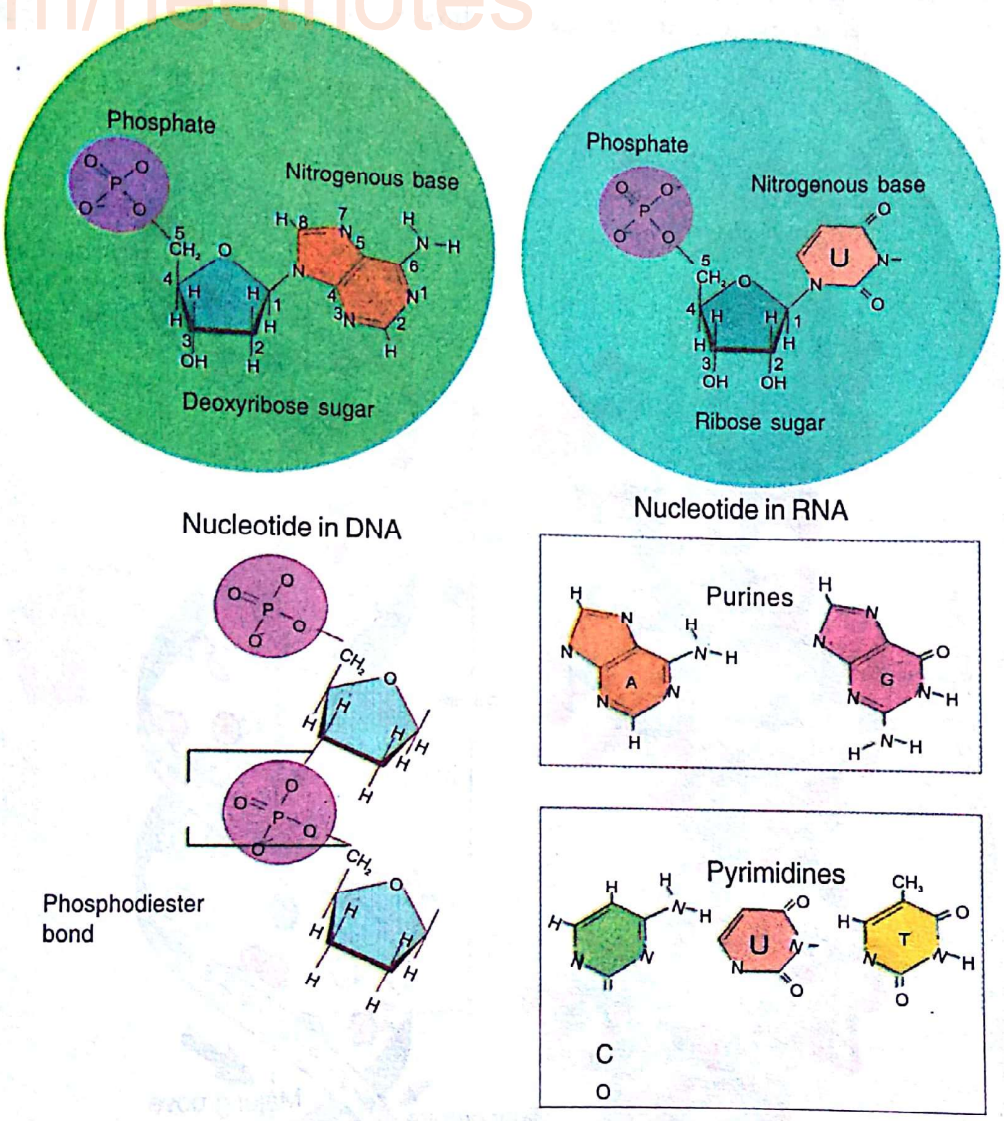

Fig. 10.9 Building blocks of nucleotide

each of which is composed of three smaller units; **a nitrogen base**, **a pentose** (5 carbon **sugar)** and a **phosphate**. The nitrogen base is a cyclic compound that can be any one of the two forms: **purines** (two rings) and **pyrimidines** (one ring). There are two types of **purines: adenine (A)** and **guanine (G)**, and three types of **pyrimidines: thymine (T), cytosine (C)** and **uracil (U)**. A characteristic that differentiates DNA from RNA is that DNA contains all of the nitrogen bases except **uracil**, and RNA contains all of the nitrogen bases except **thymine**. The nitrogen base is covalently bonded to the sugar ribose in RNA, and to deoxyribose (it has one oxygen less than ribose) in DNA. Phosphate (PO_4^{3-}), a derivative of phosphoric acid, provides the final covalent bond that connects sugar molecules in the series. Thus, the backbone of a nucleic acid strand is a chain of alternating phosphate-sugar-phosphate-sugar molecules.

The Double Helix of DNA

DNA is a long molecule formed by two long polynucleotide strands held together by hydrogen bonds. These bonds occur between complementry pairs of nitrogen bases (Fig.10.10).

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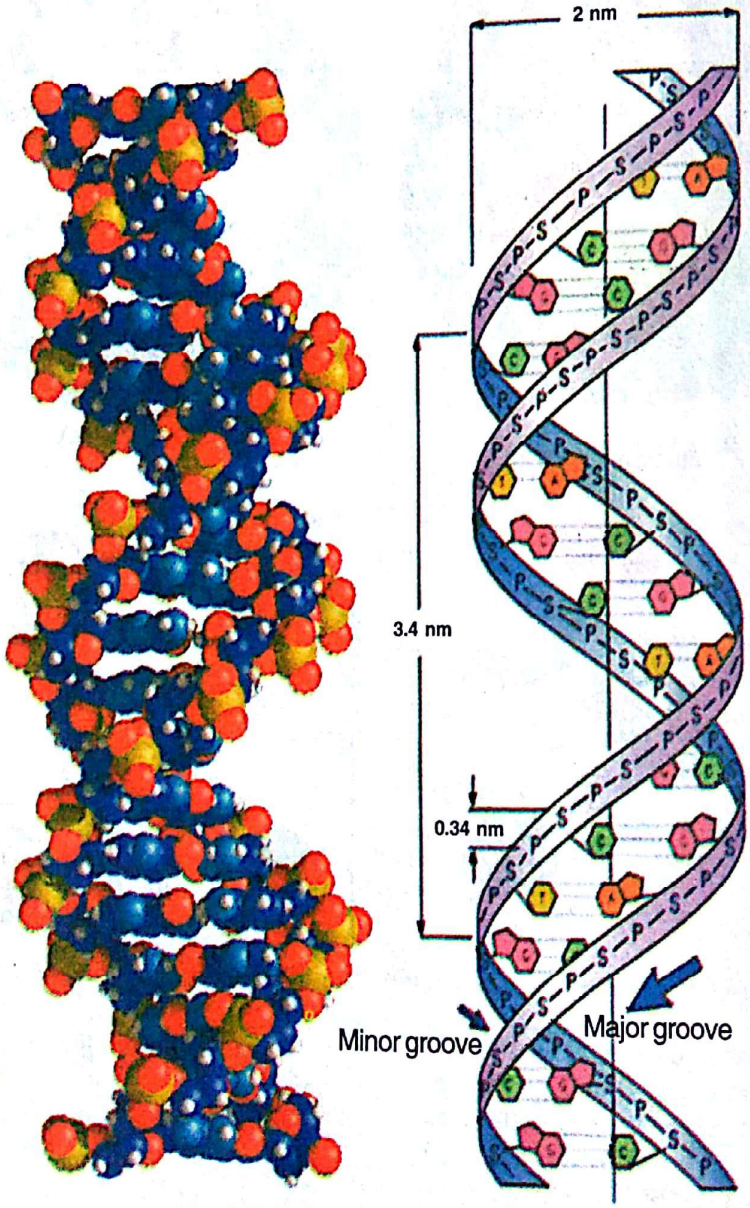

Fig. 10.10 *Double helix DNA structure*

The pairing between nitrogen bases occur according to a predictable pattern: Adenine pairs with thymine by two hydrogen bonds (A = T) and cytosine with guanine by three hydrogen bonds (C ≡ G). The DNA strands, sometimes, are compared to a twisted ladder, with the sugar-phosphate backbone representing the rails and the paired nitrogen bases representing the steps. Due to the manner of nucleotide pairing and stacking of the bases, the actual configuration of DNA is a double helix that looks somewhat like a spiral staircase. The structure of DNA is intimately related to its function.

RNA

Like DNA, RNA consists of a long chain of nucleotides. However, RNA usually has a single strand containing ribose sugar, and uracil instead of thymine. The three major types of RNA are important for protein synthesis. **Messenger RNA (mRNA)** is a copy of gene (DNA) giving the sequence of different amino acids to be incorporated into a protein; **Transfer RNA (tRNA)** is a carrier that delivers the correct amino acids for protein assembly; and **ribosomal RNA (rRNA)** is a major component of ribosomes. More information on these important processes is presented in chapter 14.

ATP

ATP is a nucleotide containing adenine, ribose and three phosphates, and is the **energy molecule of the cell**. It belongs to a category of high-energy compounds that release energy when the bond between the second and the third PO_4 is broken (Fig. 10.11). The presence of these high energy bonds makes it possible for ATP to store and release energy for cellular chemical reactions. Hydrolytic cleavage of the third phosphate not only releases energy to do cellular work but also generates adenosine diphosphate (ADP). ADP can be converted back to ATP when the third phosphate is restored, thereby serving as an energy depot or energy currency.

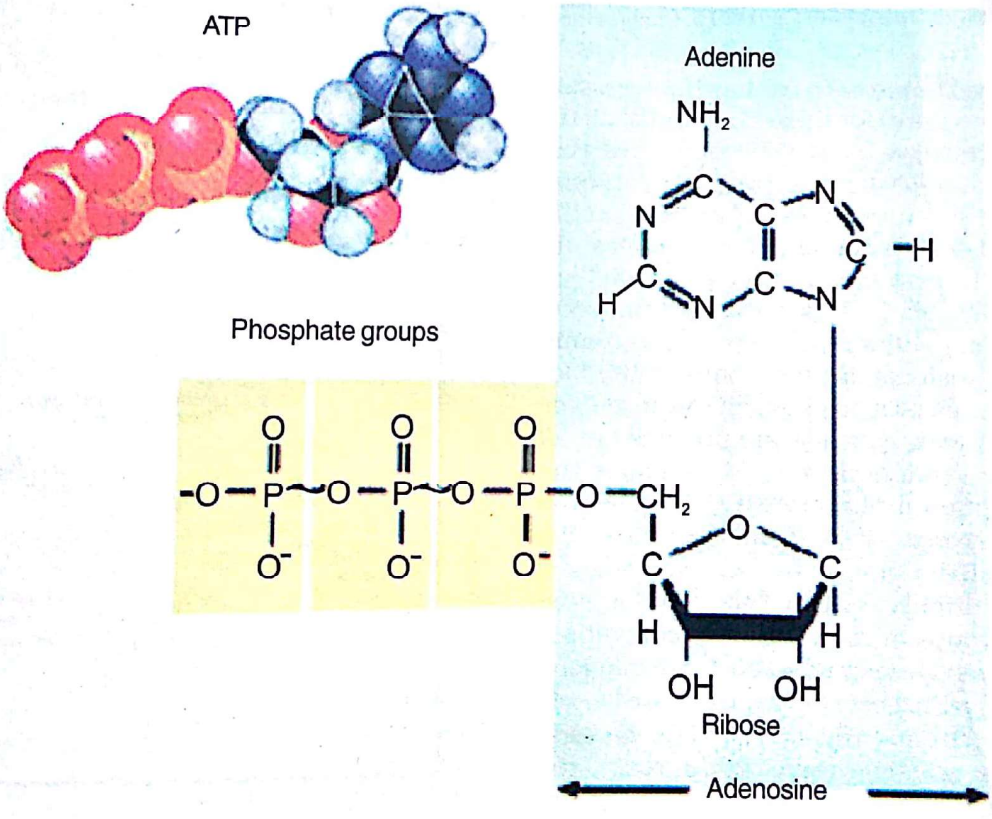

Fig. 10.11 Adenosine triphosphate – high energy molecule

Enzymes

Sometimes a small amount of a substance significantly influences the rate of a chemical reaction. The substance itself is not used up or changed at the end of the reaction. Such a substance is called a **catalyst**, and the phenomenon **catalysis**. The latter may be positive if the reaction rate is enhanced, or negative if the reaction rate is reduced. Both organic and inorganic substances can play the role of catalysts. The rate and efficiency of the reactions in a cell depend on certain special molecules called **Enzymes** which are synthesised by the cells. Enzymes are biological catalysts. The DNA in each cell has the necessary message (blue print) for the production of all the enzymes required by it. The cell uses this information as and when necessary to synthesize the enzymes required to catalyse specific reactions at any point of time. Enzymes are synthesized by living cells. But they retain their catalytic ability even when extracted from cells. Rennet tablets containing the enzyme rennin from the calf's stomach have long been used for coagulating milk protein to obtain caesin (cheese from milk). Mostly enzymes are proteins but all proteins are not enzymes. Majority of the enzymes contain a non-protein part called the **Prosthetic group**. It is tightly bound to the enzyme. Prosthetic groups are metal compounds. For example, iron-porphyrin complexes form the prosthetic groups of cytochromes. In addition, certain organic compounds and inorganic ions are required for enzyme activity. They are loosely bound to the enzyme and are called **Co-factors**. Nicotinamide adenine dinucleotide (NAD), the precursor of nicotinic acid (niacin), and flavin adenine dinucleotide (FAD), the active form of vitamin B_2 (riboflavin) are organic co-factors or **Coenzymes** of many oxidising enzymes of the mitochondria. Certain metals, especially those occurring in trace amounts also facilitate enzyme reaction. Iron (Fe^{++}) is a co-factor responsible for the catalytic action of catalase.

The three-dimensional structure of enzyme has a specific catalytic site called the **Active site**. An enzyme may have more than one active site. has a specific catalytic site called The active site serves as a 'lock' into which the reactant (commonly referred to as the **substrate**) fits in like a key (Fig. 10.12). The point where the substrate is bound on the active site is known as the substrate-binding site. Substrate binding causes a lowering of the activation energy and allows the reaction to proceed (Fig. 10.13). Once the reaction is completed, the enzyme releases the **products** and is ready to catalyse again. How fast do enzymes act?

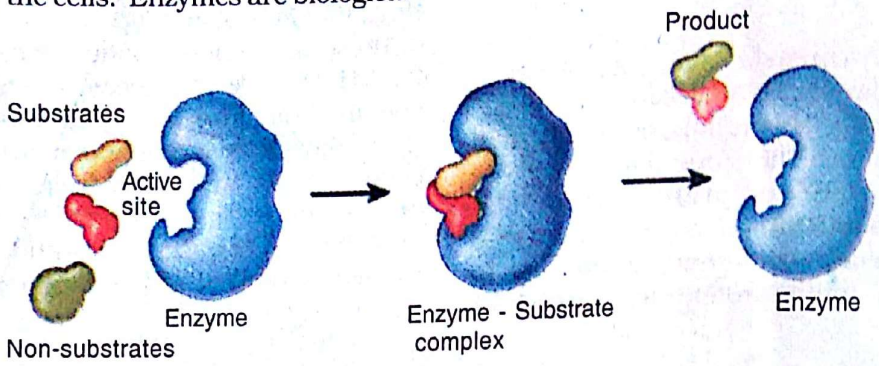

Fig. 10.12 Enzyme action – lock and key mechanism

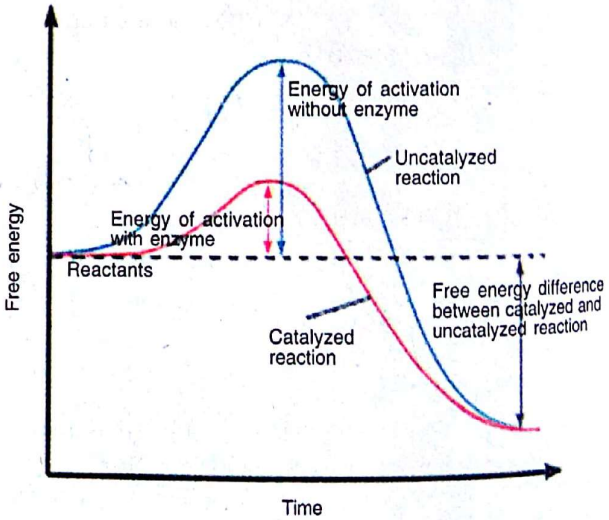

Fig. 10.13 Activation energy

A single molecule of the enzyme carbonic anhydrase, the fastest enzyme known, hydrates 36 million (36×10^6) molecules of carbon dioxide per minute

$$CO_2 + H_2O \xrightarrow{\text{Carbonic anhydrase}} H_2CO_3$$

The catalysed reaction is 10 million times faster than the non-catalysed reaction.

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Properties of Enzymes

Specificity

Each enzyme can catalyse the change of either a specific substrate or a specific group of substrates.

Optimum Temperature

Enzymes generally function in a narrow range of temperature which usually corresponds to the body temperature of the organism. Each enzyme shows its highest activity at a particular temperature called optimum temperature. Activity declines both above and below the optimum temperature. Low temperature preserves the enzyme in a temporarily inactive state. Food may be preserved for a long time in a frozen state because neither microbial enzymes nor enzymes in the food can act at low temperatures to cause its spoilage. High temperature destroys enzyme activity, because proteins are denatured by heat. For this reason, only a few cells can tolerate temperatures above 45°C. Some heat resistant microorganisms living in hot springs at temperatures close to 100°C, possess heat-resistant enzymes.

Optimum pH

Each enzyme shows its highest activity at a specific pH. This is called the optimum pH. Activity declines both above and below the optimum pH. Most intracellular enzymes function best around neutral pH. Some digestive enzymes have their optimum in the acidic or alkaline range. For example, the protein digesting enzyme pepsin found in the stomach, has an optimum pH of 2.0. Another protein-digesting enzyme, trypsin, found in the duodenum, functions best in an alkaline pH of 8.5.

Enzyme-Substrate Complex

Each enzyme (E) has a substrate-binding site in its molecule. A highly reactive enzyme-substrate complex is consequently produced. The latter almost immediately dissociates into the product or products (P) and the unchanged enzyme.

$$E + S \rightleftharpoons ES \longrightarrow EP \longrightarrow E + P$$

(Short-lived)

Formation of the enzyme-substrate complex is essential for catalysis. The higher the affinity of the enzyme for its substrate, the greater is its catalytic activity.

Effect of substrate concentration

With the increase in substrate concentration (S), the velocity V of the enzymatic reaction rises at first. The reaction ultimately reaches a maximum velocity(Vmax), which is not exceeded by any further rise in substrate concentration (Fig. 10.14). This happens because enzyme molecules are fewer than the substrate molecules. Any more increase in susbtrate concentration will saturate all the enzyme molecules. No enzyme is left free to bind with additional molecules of the substrate.

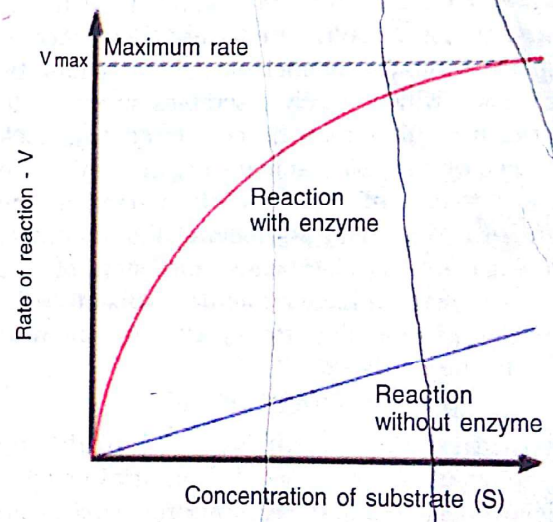

Fig.10.14 *Effect of substrate concentration on enzyme action*

Inhibition of Enzyme Action

Enzyme action can be inhibited in four different ways. Inhibition of enzyme action by denaturation of proteins has already been

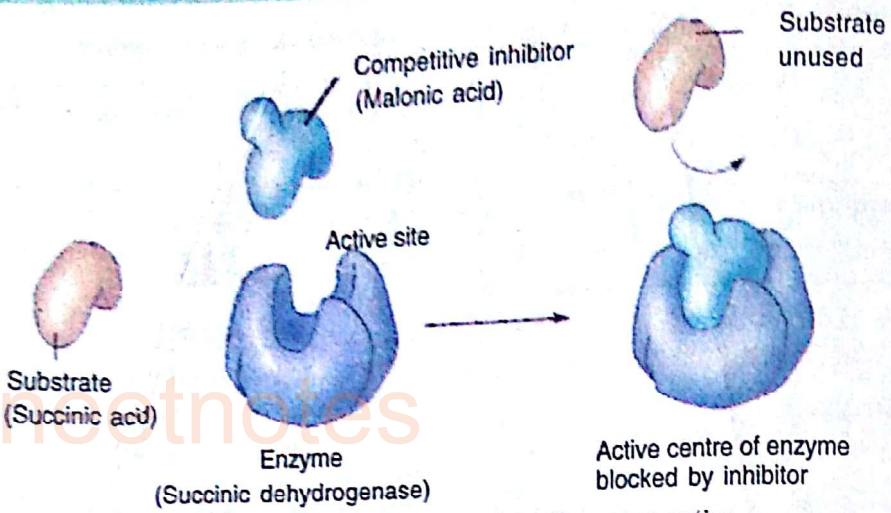

Fig. 10.15 Competitive inhibition of enzyme action

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mentioned. The other three ways in which enzymes can be inhibited are:

Competitive Inhibition

The action of an enzyme may be reduced or inhibited in the presence of a substance which closely resembles the substrate in its molecular structure. Such an inhibitor is called a **competitive inhibitor** of that enzyme. Due to its close structural similarity with the substrate, the inhibitor competes with the latter for the substrate-binding site of the enzyme (Fig. 10.15). Consequently, the enzyme cannot bind to the substrate. As a result, the enzyme action declines, e.g., the inhibition of succinic dehydrogenase by malonate, which closely resembles succinate in structure. This may be compared to a lock jammed by a key similar to the original key. Such competitive inhibitors are often used in the control of bacterial pathogens. For instance, sulpha drugs are competitive inhibitors of folic acid synthesis in bacteria as they substitute for p-amino benzoic acid, thus preventing the next step in the synthesis.

Non-competitive Inhibition

Cyanide kills an animal by inhibiting cytochrome oxidase, a mitochondrial enzyme essential for cellular respiration. This is an example of non-competitive inhibition of an enzyme. Here the inhibitor (cyanide) has no structural similarity with the substrate (cytochrome c) and does not bind with the substrate-binding site but binds at some other site of the enzyme and destroys the catalytic function of the enzyme. Thus in non-competitive inhibition substrate binding takes place but no products are formed.

Allosteric Modulation or Feedback Inhibition

The activities of some enzymes, particularly those which form a part of a chain of reactions (metabolic pathway), are regulated internally (see Fig 2.10). Some specific low molecular weight substances, such as the product(s) of another enzyme in the chain, acts as the inhibitor. Such a modulator substance binds with a specific site of the enzyme different from its substrate-binding site temporarily. This binding increases or decreases the enzyme action. Such enzymes are called **allosteric enzymes**; e.g. hexokinase which changes glucose to glucose-6-phosphate in glycolysis. Decline in enzyme activity by the allosteric effect of the product is called **feedback inhibition**, e.g., allosteric inhibition of hexokinase by glucose-6-phosphate (Fig. 10.16).

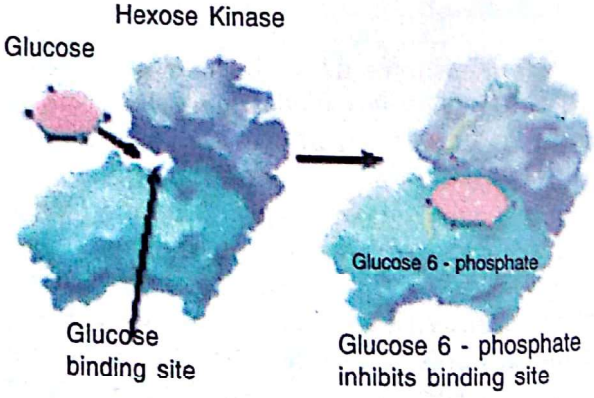

Fig. 10.16 Feedback inhibition

Hormones

The word hormone has been derived from a Greek word (*hormòn*) meaning 'to arouse to activity'. By the classic definition, a hormone is a substance that is synthesised in minute quantities in one tissue and transported by circulatory system to another organ. The tissue or organ where they are produced are called **effectors** and those where they exert their influence are called **targets**. Based on their site of action hormones are of two types: local and general. The local hormones have specific local effects, for example, cholecystokinin. On the other hand, the general hormones are secreted by various endocrine glands and are transported through the blood to cause physiological action at points away from their place of origin, e.g. growth hormones, thyroid hormone, adrenocorticotropin, etc.

The hormones are required in extremely small quantities and perform a variety of regulatory functions ranging from growth, vegetative and sexual development, cellular oxidation to thermal regulation and the metabolism of carbohydrates, proteins and fats. Hormone action at the cellular level begins by its association with specific receptor.

The plant hormones are known as 'phytohormones'. These are organic compounds produced naturally in higher plants and controlling growth or other physiological function either at the site of their origin or far remote from their place of production. Auxins, gibberellins, cytokinins, abscisic acid (ABA), and ethylene are the five major types of hormones found in plants.

Vitamins

These are organic molecules in food that are required in minute quantities for normal metabolism but cannot be synthesised in adequate amounts by humans and animals. A dietary or physiologic deficiency of anyone of them leads to a specific set of disease symptoms that can be corrected by administration of that vitamin alone. The vitamins are synthesised by plants and bacteria. These are classified on the basis of solubility as:

(a) **Water Soluble Vitamins** which include the B-complex group of vitamins, and vitamin C (ascorbic acid). B-complex vitamins are found in whole grain cereals, legumes, leafy green vegetables, meat and dairy products. Citrus fruits are good source of vitamin C.

(b) **Fat Soluble vitamins** are soluble in fats, e.g. vitamin A, D, E and K. These are present in food fats, e.g. fatty meats, liver, dairy fats, yolks, vegetable seed oils, etc. The vitamins function as coenzyme or cofactor and are required in very small quantities for normal metabolism of animals including us.

SUMMARY

The common inorganic materials such as salts, minerals and water are present in the cells. Minerals are grouped as major and minor based on their amount required by the cells. These are the constituents of cellular components like proteins, amino acids and also bones and teeth. The deficiency of minerals results in disorders. Water is an important constituent of cells. It is a good solvent and helps in maintaining all the chemical reactions and molecular conformations. It is also helpful in maintaining the constancy of internal environment at the cellular level. Biochemical macromolecules of the cell are categorised into four main classes: Carbohydrate, lipids, proteins and nucleic acids. Basic unit of carbohydrate is a monosaccharide which is a polyhydroxy aldehyde or ketone molecule containing 3-7 carbons, a disaccharide is combination of two and polysaccharide is combination of many monosaccharides. The three main types of polysaccharides are cellulose-structural component of cell wall, starch-storage product in plants, and glycogen-storage product in animals. Other structural polysaccharides are agar, chitin, peptidoglycan, lipopolysaccharide and, glycocalyx. Some of them act as recepters in cells. Lipids are long chain of fatty acid bonding with glycerol, major types are sterols, triglycerides, phospholipids and waxes. Triglycerides are of two types: saturated and unsaturated. Phospholipids are

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structural components of cell membrane. They have hydrophobic or hydrophilic ends. The sterol called cholesterol reinforces the structure of the cell membrane in animals and wall-less bacteria. Wax, an ester of long chain alcohol with saturated fatty acid, provides extra protection to cells by making them water proof. Proteins are a long chains of amino acids. There are 20 amino acids in the living world. All proteins are products of various combinations of these amino acids. A protein has primary, secondary and tertiary structure by folding of peptide chain at specific points. This gives a protein three-dimensional structure. Some antibodies or enzymes fold another time and form quaternary structure also. The disruption in structure results in denaturation of proteins.

The nucleic acids are of two kinds: DNA and RNA. They contain special coded genetic programme with detailed and specific instructions for controlling cell activity and organism's heredity. Nucleic acids are polymers of nucleiotides. Each nucleotide is made of a nitrogen base, pentose sugar-deoxyribose in DNA and ribose in RNA, and a phosphate. The backbone of a nucleic acid is a chain of alternating phosphate-sugar-phosphate sugar molecules. DNA is formed by two long polynucleotide strands held together by hydrogen bonds. The structure is then helically twisted around an axis. RNA is similar as DNA but it is single stranded. RNA has uracil instead of thymine nitrogen base. It is of three types messenger RNA, transfer RNA and ribosomal RNA . It participates in protein synthesis and acts as the genetic material in organisms where DNA is absent. ATP is also a nucleotide containing the nitrogenous base adenine, ribose sugar and three phosphates. It has three energy rich bonds which store energy and release when broken down.

Enzymes are chemical substances which catalyse the reactions in the cells. The enzymes are secreted and synthesised by the living cells. Most enzymes are proteins but all proteins are not enzymes. Most enzymes contain a non-protein part called prosthetic group. The enzyme action is facilitated by some organic or inorganic ions and certain metals. Enzymes generally exhibit substrate specificity, require optimum temperature and pH for activity, form a short-lived enzyme-substrate complex and are affected by substrate concentration. The enzymatic activity can be inhibited by competitive, non-competitive or feedback inhibitors.

The chemical substances called hormones are synthesized both by plants and animals. The place of action of hormones is frequently different from the place of synthesis. These are required in small quantities and control many physiological processes. Another group of organic substances called vitamins are also required in small amounts. These are mainly synthesised in plants. Vitamins are either fat soluble or water soluble.

EXERCISES

1. What are macromolecules? Give examples.
2. Give two examples of storage polysaccharide.
3. What is chitin?
4. Explain how glycosidic bonds are formed.
5. Describe functions of polysaccharides in living organisms.
6. What is meant by tertiary structure of proteins ?
7. Mention the difference between saturated and unsaturated fat?
8. Explain the composition of triglyceride.
9. Describe the structure of phospholipid. How are they arranged in the cell membrane?
10. Write short notes on
    (i) steroids
    (ii) wax

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CHAPTER 11

CELL CYCLE

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You are familiar with cell and its various organelles, which enable the cells to perform different functions. The cells arise by the division of pre-existing cells and undergo differentiation. You will study more on some of these aspects in this chapter.

The importance of cell division can be appreciated by realising that in higher eukaryotes life starts with the formation of zygote (fertilised egg). From this single cell, trillions of cells develop by successive divisions. These cells undergo the process of differentiation and organs are formed. During this, the rate of cell division and the number of dividing cells vary. There are fewer cells dividing in a fully-formed organism than during the course of development. However, in adult tissues like bone marrow, germinal tissues (in animal) and meristematic regions (in plants), cell division takes place continuously.

11.1 CELL CYCLE

When a cell is to divide, it is expected to synthesise its various components including the genetic material so that all materials get duplicated. After division the duplicated material passes into the daughter cells. *This orderly sequence of events by which the cell duplicates its contents and then divides into two is termed as cell cycle.* The events that control the above sequence of activities are genetically controlled.

The cell cycle comprises essentially of two stages.

(i) Interphase, and
(ii Mitosis.

Cell, however, spends most of its life-span in interphase. The interphase as is known today is a period of intense biosynthetic activity in which the cell doubles in size and duplicates precisely its chromosome complement. The nerve cells of mammals, however, do not divide at all after birth. Thus, for a human neuron the interphase period lasts the entire life of a person. In eukaryotes, the division generally takes place by mitosis or meiosis. The period of division represents a complex series of stages by which cellular material is divided equally between the daughter cells. Cell division is only the final phase. Before the cell enters cell division, it has already duplicated its molecular components. In this respect, cell division can be considered as *the final separation of the already duplicated molecular units.*

Interphase

Though this phase is sometimes called 'resting stage', but it is in fact a period of great activity. Three important processes, which are preparatory to cell division take place during interphase. These processes are:

(i) Replication of DNA along with the synthesis of nuclear proteins such as the histones.
(ii) In animal cells, duplication of a centriole takes place by the assembly of daughter centrioles close to the parent centrioles, which are at right angle to each other.
(iii) Synthesis of energy-rich compounds, which provide energy for mitosis, and synthesis of proteins at the end of interphase.

The interphase can be divided into three phases:

(i) The post mitotic gap phase **(G_1)** takes place at the end of one cell division. RNA and protein are synthesised during this period, but there is no synthesis of DNA.
(ii) During the synthesis phase **(S)**, DNA is replicated and the DNA content of the nucleus is doubled.

(iii) During the pre-mitotic gap phase (G_2), synthesis of RNA and protein continues, but DNA synthesis stops. The durations of the S phase, the G_2 phase and mitosis is generally constant in most cell types.

The length of G_1 phase is usually variable. Cells that do not divide frequently have a longer G_1 phase, while frequently dividing cells have a shorter G_1 phase. During G_1 phase, a cell may follow one of the three options: (a) cell may continue on the cycle and divide, (b) the cell can permanently stop division and enter G_0 or quiescent stage, and (c) the cell cycle may be arrested at a specific point of G_1 phase. The cell in the arrested condition is said to be in the G_0 state. The cell in the G_0 may be considered to be withdrawn from the cell cycle. When conditions change and growth is resumed the cell re-enters the G_1 period. Eukaryotic chromosomes undergo condensation–decondensation cycle during interphase. G_1 chromosomes are completely dispersed.

In many organisms most important point in the regulation of the cell cycle occurs in the G_1 phase, during which it must decide whether the cell will start a new cycle or will become arrested in G_0 phase. Once this G_1 check point has been passed, the cell goes on to complete a new cycle.

The G_1 phase can be terminated by various stimuli, and cell division then starts. Once a higher eukaryotic cell entered the S phase and has begun DNA replication, it has usually committed itself to division. During interphase, replications of chromosomes takes place so that each chromosome now consists of two chromatids. Following this, the cell enters into the mitosis (M) phase. Hence, the cell cycle is divided into four phases : G_1 phase, S phase, G_2 phase and M phase (Fig. 11.1).

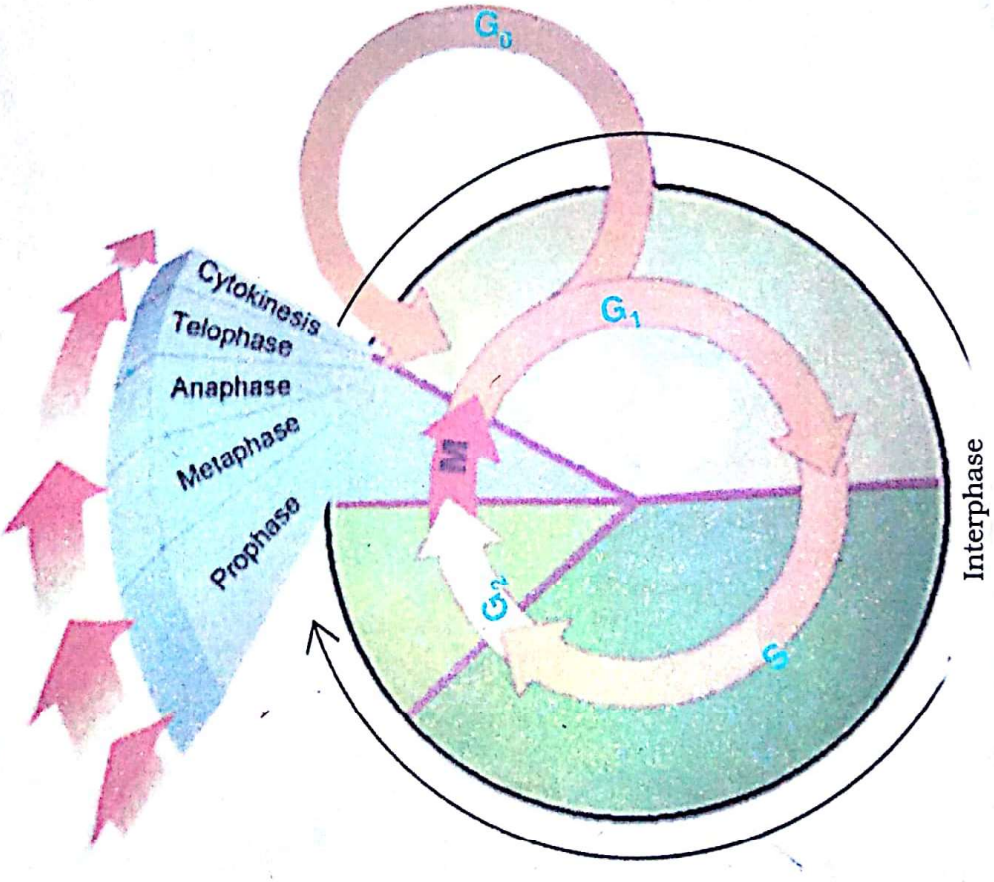

Fig. 11.1 Phases of cell cycle

11.2 MITOSIS

The process of cell division whereby the chromosomes are duplicated and distributed equally to the daughter cells is called **mitosis**. It is also called equational division. The mitotic cycle is divided into four phases: **Prophase, Metaphase, Anaphase and Telophase** (Fig. 11.2).

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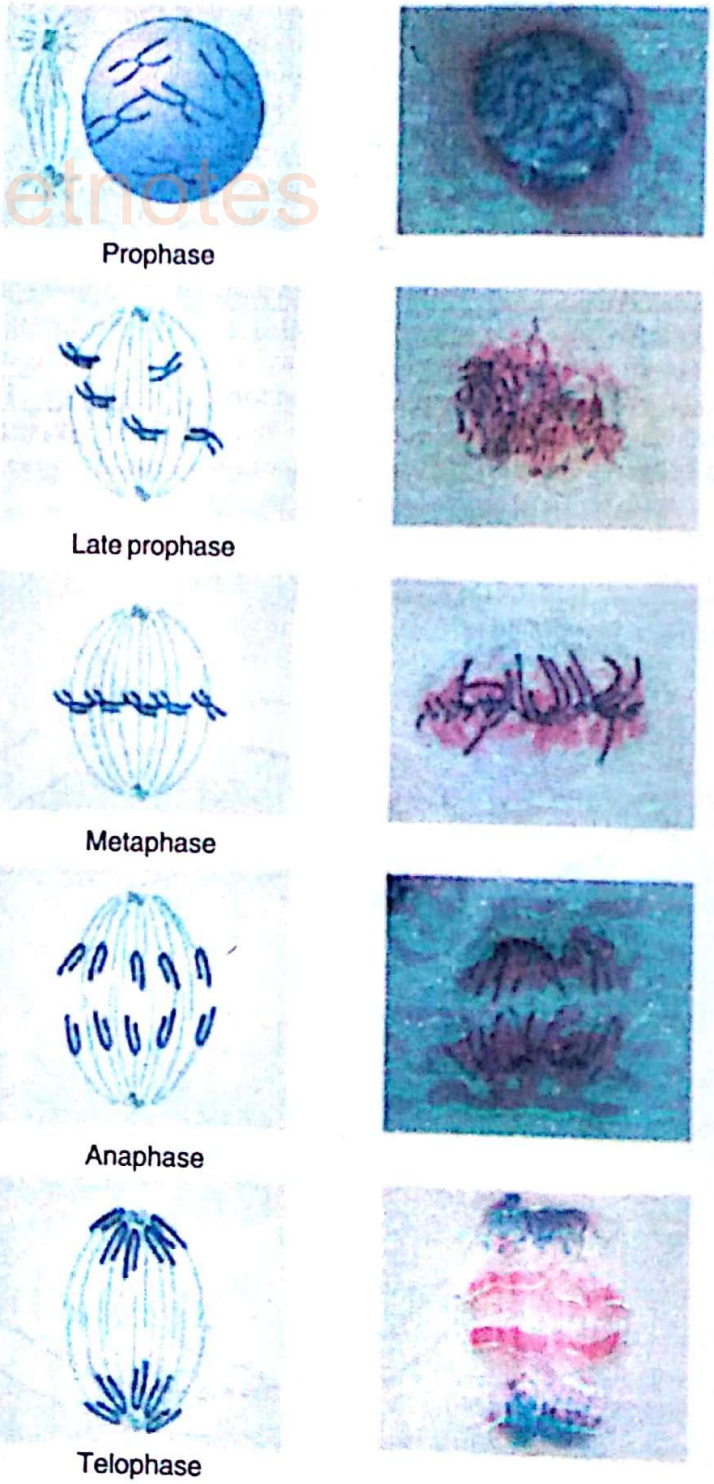

Fig. 11.2 Mitosis – different stages in cell – diagramatic (left) microphotograph (right)

Prophase

During prophase the cell nucleus becomes spheroid, and there is an increase in viscosity of cytoplasm. The chromosomes shorten and thicken and become stainable. By the end of prophase some chromosomes may contract upto 1/25th of their length in early prophase. The double-stranded nature of the chromosomes is now visible. Each chromosome consists of two chromatids called sister chromatids held together by a centromere. With the progress of prophase, the chromosomes which were essentially distributed randomly during prophase migrate towards the nuclear membrane.

The centrosome, which had undergone duplication during interphase, now begins to move towards opposite poles of the cell (Fig. 11.3). Centriole with radiating microtubules is called aster (star-like). The aster together with cytoplasmic spindle is called mitotic apparatus. The spindle begins to be formed between the two poles. The spindle consists of microtubules that are made of the proteins called tubulins and proteins associated with them. The spindle is a dynamic structure, and undergoes a cycle of dissolution and reformation.

Metaphase

The beginning of prometaphase is marked by the disappearance of the nuclear membrane. When the nuclear membrane dissolves, there is no differentiation between cytoplasm and nucleoplasm. The chromosomes are attached to the spindles through their centromeres. Such a mitosis is called extra-nuclear mitosis or **eumitosis**. In many protozoans and some animals cells, however, the nuclear membrane does not disappear during cell division. The mitosis takes place within the nuclear membrane and is called **intranuclear mitosis** or **premitosis**. The chromosomes move freely and proceed towards the equator.

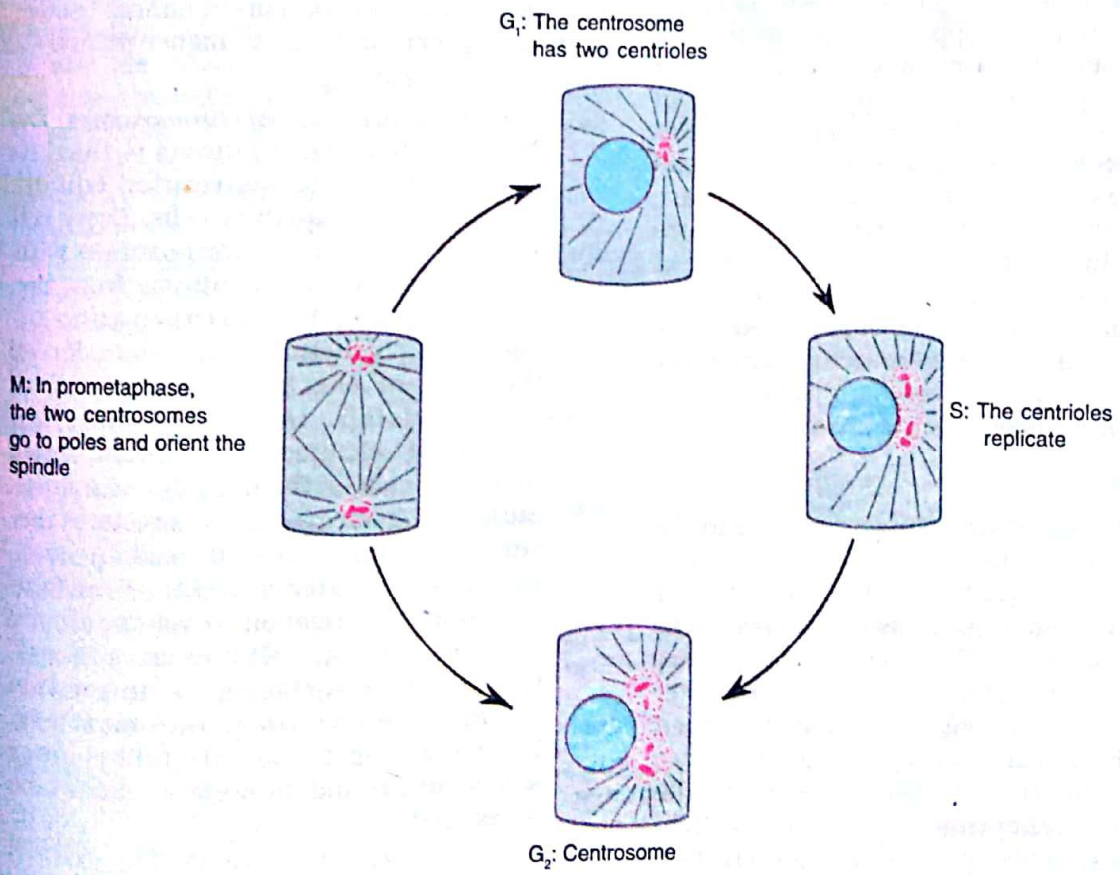

Fig. 11.3 Centrosome duplication during prophase

Later in metaphase, the chromosomes usually are lined up in one plane to form the **equatorial plate** or **metaphasic plate**. Occasionally, only the centromere lies on the equatorial plane, while the chromosome arms are directed away from the equator. Smaller chromosomes are usually central in position whereas the larger ones are peripheral (Fig. 11.2).

Anaphase

The chromosomes are arranged on the equatorial plate for a short period only. The sister chromatids begin to separate from each other, starting at the centromere. They are now called daughter chromosomes. These now behave as if they repel each other. The two sets of chromosomes migrate towards the poles. The chromosome movement is brought about by the shortening of spindle fibres attached to the centromeres.

Telophase

Telophase begins when the two sets of daughter chromosomes reach opposite poles of the cell and the spindle disappears. A new nuclear membrane is formed around each set of chromosomes. The nucleoli reappear at regions called **nucleolar organisers**, in one or more pairs of chromosomes. Telophase changes are associated with the restoration of the interphase condition. In a way, telophase is the reverse of prophase. Each daughter cell gets the same complement of chromosomes and nucleoli as of the mother cell. The chromosomes gradually uncoil and become less compact. They eventually lose their staining ability.

Cytokinesis

In animal cells a cleavage furrow appears in the plasma membrane at the equator at the beginning of telophase. This furrow or constriction becomes progressively deeper as the spindle breaks down. Eventually, the ingrowing constrictions join and separate the two daughter cells. This division of cytoplasm is called **cytokinesis**. When nuclear division takes place without cytoplasmic division it results in the formation of a **syncytium**, which is a condition where large number of nuclei are present in a single cell.

In plants, there is a formation of cell plate between the two daughter nuclei. This grows from the middle towards the periphery, and finally joins the cell wall. The cell plate represents the middle lamella between the walls of two adjacent cells.

During division, cell organelles like mitochondria, plastids, Golgi complex, lysosomes and the cytoplasmic matrix are distributed into the two daughter cells. Of these, mitochondria and plastids reproduce themselves, but details of the fate of other organelles are not yet known.

You should not confuse nuclear division in mitosis with that in amitosis. Fleming in 1882 described **amitosis**. The nuclear division in amitosis occurs by a process other than mitosis. A dumbbell shaped cleavage of the cell nucleus occurs during which chromosomes are not recognisable and spindle is not formed. Amitosis may or may not be followed by the division of the cell, and nuclei so formed are frequently of unequal size. This process occurs in certain protists, ciliates, in specialised animal tissues, and old degenerating cells of higher plants.

Significance of Mitosis

(i) *Equal distribution of chromosomes:* The essential feature of mitosis is that the chromosomes are distributed equally among the two daughter cells. Every cell involves division of chromosomes with repeated divisions by mitosis from the zygote onwards, maintenance of identical genetic constituents for all the cells of the body is ensured at each division. Hence the constant number of chromosomes is maintained in all the cells of the body due to mitosis.

(ii) *Surface/Volume ratio:* Mitosis restores the surface/volume ratio of the cell. A small cell has a greater amount of surface available in relation to volume than a large cell. As the cell increases in size, the available surface area, in relation to increased volume, becomes less. By undergoing division, the cellbecomes smaller in size and the surface volume ratio is restored.

(iii) *Nucleo- cytoplasmic ratio:* The growth of multicellular organisms is due to mitosis. A cell cannot grow in size to a large extent

without disturbing the ratio between the nucleus and the cytoplasm. After a particular size has been reached, the cell divides to restore the nucleo-cytoplasmic ratio. Thus growth takes place mainly by an increase in the number of cells . An idea of growth due to mitosis can be obtained from the fact that the human body contains about 6 x 10^{12} cells, all are ultimately derived by the division of a single zygote cell.

(iv) Repair: Repair of the body takes place because of the addition of cells by mitosis. The cells of the upper layer of the epidermis, cells of the lining of the gut, and RBCs are constantly being replaced. It is estimated that in the human body about 5 x 10^9 cells are lost and replaced daily.

11.3 MEIOSIS

Meiosis occurs in the germ cells, which are destined those to form gametes in sexually reproducing organisms. Many stages of meiotic division are similar to those of mitosis. Meiosis comprises **meiosis I** and **meiosis II**. Each one of these includes prophase, metaphase, anaphase and telophase. During the first meiotic divison, the members of each homologous pair of chromosomes separate and are distributed into separate cells. In the second meiotic division, the sister chromatids of each chromosome separate and are distributed to the daughter cells. In this way the number of chromosomes per cell is ultimately reduced by half.

The meiotic division takes place at the end of the G_2 phase of the interphase, as in the case of mitotic cell division. The essential events that take place during meiosis are:

(i) Two successive divisions without any DNA replication occurring between them,
(ii) Pairing and formation of chiasmata and crossing over,
(iii) Segregation of homologous chromosomes, and
(iv) Separation of sister chromatids.

Prophase I

From the morphological point of view, the prophase of the first meiotic division (Prophase I) is a long process during which homologous chromosomes pair closely and interchange hereditary material. For convenience, the first meiotic prophase is divided into the following five sub-stages: **Leptotene** (leptonema), **Zygotene** (Zygonema), **Pachytene** (Pachynema), **Diplotene** (Diplonema), and **Diakinesis.**

Now we will study about each of these stages in some detail.

Leptotene

The leptotene stage begins when each chromosome is first seen to have condensed from its interphase conformation to produce a long thread. Each chromosome is attached at both of its end to the nuclear envelop via a specialised structure called **attachment plate**. Although each chromosome has replicated and consists of two sister chromatids, these chromatids are very close to each other and therefore chromosomes appear to be single-stranded (Fig. 11.4a).

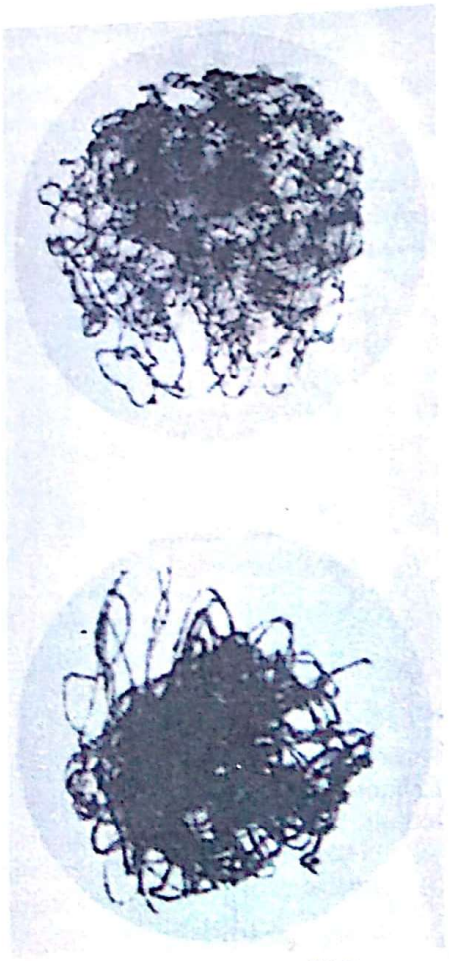

Fig. 11.4 (a) Leptonema (b) Zygonema

Zygotene

Zygotene stage begins as soon as intimate pairing between the two members of each homologous chromosome pair is initiated by the process called **synapsis** or zygotene pairing. The paired homologous chromosomes comprise one chromosome derived from each parent. Synapsis often starts when the homologous ends of the two chromosomes are brought together on the nuclear envelope. It continues inwards in a zipper-like manner from both ends aligning the two homologous chromosomes side by side. The pairing is completed in three different ways as follows:

(i) Proterminal Pairing: The two homologous chromosomes start pairing at the terminals, which gradually progresses towards the centromere.

(ii) Procentric Pairing: The pairing starts at the centromere and proceeds towards the end.

(iii) Random or Intermediate Pairing: The pairing may be at many points towards the ends.

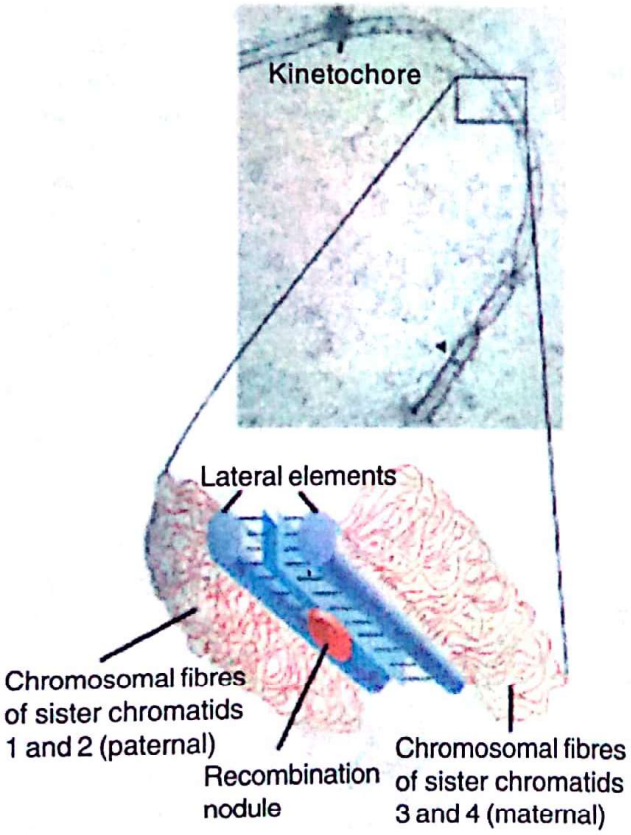

Fig. 11.5 Synaptonemal complex

As a result of synapsis each gene is brought into close contact with its allele located on the homologous chromosome. The two homologous chromosomes are brought together through a characteristic ladder-like structure, called **synaptonemal complex** (Fig. 11.5).

The chromosome configuration so produced is called a **bivalent**. Each of the homologous chromosomes in meiotic prophase I consists of two closely apposed sister chromatids, thus each bivalent contains four chromatids, and is also called **tetrad**.

Pachytene

As soon as the synapsis is complete all along the chromosome, the cells are said to have entered the pachytene stage of prophase, where they may remain for days. At this stage, large recombination nodules appear at intervals on the synaptonemal complex (Fig. 11.5). These recombination nodules are thought to mediate for chromosomal recombination. The non-sister chromatids (chromatids of different chromosomes of a homologous pair) exchange segments with each other as a result of crossing over.

Diplotene

The beginning of diplotene stage is marked by the beginning of separation of the paired homologous chromosomes.The separation is however not completed. The homologous chromosomes remain attached at one or more points where crossing over has occurred. These points of attachment are called **chiasmata**. In oocytes, diplotene can last for months or years, since it is at this stage that the chromosomes decondense and engage in RNA synthesis.

Diakinesis

The beginning of diakinesis is marked by **terminalisation of chiasmata.** At this stage, RNA synthesis stops and the chromosomes condense, thicken, and become attached to the nuclear envelope. Each pair of sister chromatids is attached at their centromeres, whereas non-sister chromatids of homologous

Fig. 11.6 Diakinesis in meiosis

chromosomes are in contact with each other at or near their telomeres (Fig. 11.6).

Metaphase I

During metaphase–I, the bivalents become arranged in the plane of the equator forming the equatorial plate (Fig. 11.7). The centromere of each chromosome is directed towards the opposite poles and the arms of chromosomes face the equatorial plate.

Anaphase I

The two members of each bivalent seem to repel each other and move towards the opposite poles. Thus each pole receives half the number of chromosomes or the haploid set of the chromosomes. Thus, actual reduction in number of chromosomes occurs. The movement of chromosomes is brought about by the shortening of spindle fibres, similar to that in during mitosis.

Telophase I

The nuclear membranes are formed during this stage by the endoplasmic reticulum around the groups of daughter chromosomes with the appearance of one nucleolus in each nucleus. It results in the formation of two daughter cells each with haploid number of chromosomes.

The Second Meiotic Division

The second meiotic division is essentially similar to mitosis. It divides each haploid meiotic nucleus into two daughter haploid nuclei. Like mitotic division it can be described under four phases (Fig. 11.7):

Prophase II

Prophase II does not show the complex nuclear behavior of prophase I but conforms to the features of mitotic prophase. Spindle formation takes place in prophase II as in mitosis, and the nuclear membrane disappears.

Metaphase II

The chromosomes become oriented on the equatorial plate and have the same relationship to the spindle as in mitosis.

Anaphase II

The centromere divides and the two sister chromatids of each chromosome separate and move towards the poles. After separation, each chromatid behaves as a chromosome. Thus, a chromosome has one chromatid before and two chromatids after replication.

Telophase II

At this stage, the four groups of chromosomes become organised into four haploid nuclei. The chromosomes return to the interphase condition. The endoplasmic reticulum forms the nuclear envelope around the chromosomes and the nucleolus reappears due to association of rRNA with ribosomal proteins synthesised on rDNA templates. Each nucleus at this stage contains the haploid number of chromosomes and forms four cells.

Significance of Meiosis

(i) The meiosis maintains a definite and constant number of chromosomes in the sexually reproducing organisms by producing haploid gametes.

(ii) By crossing over, the meiosis provides an opportunity for genetic variation through gentic material by crossing over and random distribution of maternal and paternal chromosomes. The variation serves as the raw material for the evolutionary process.

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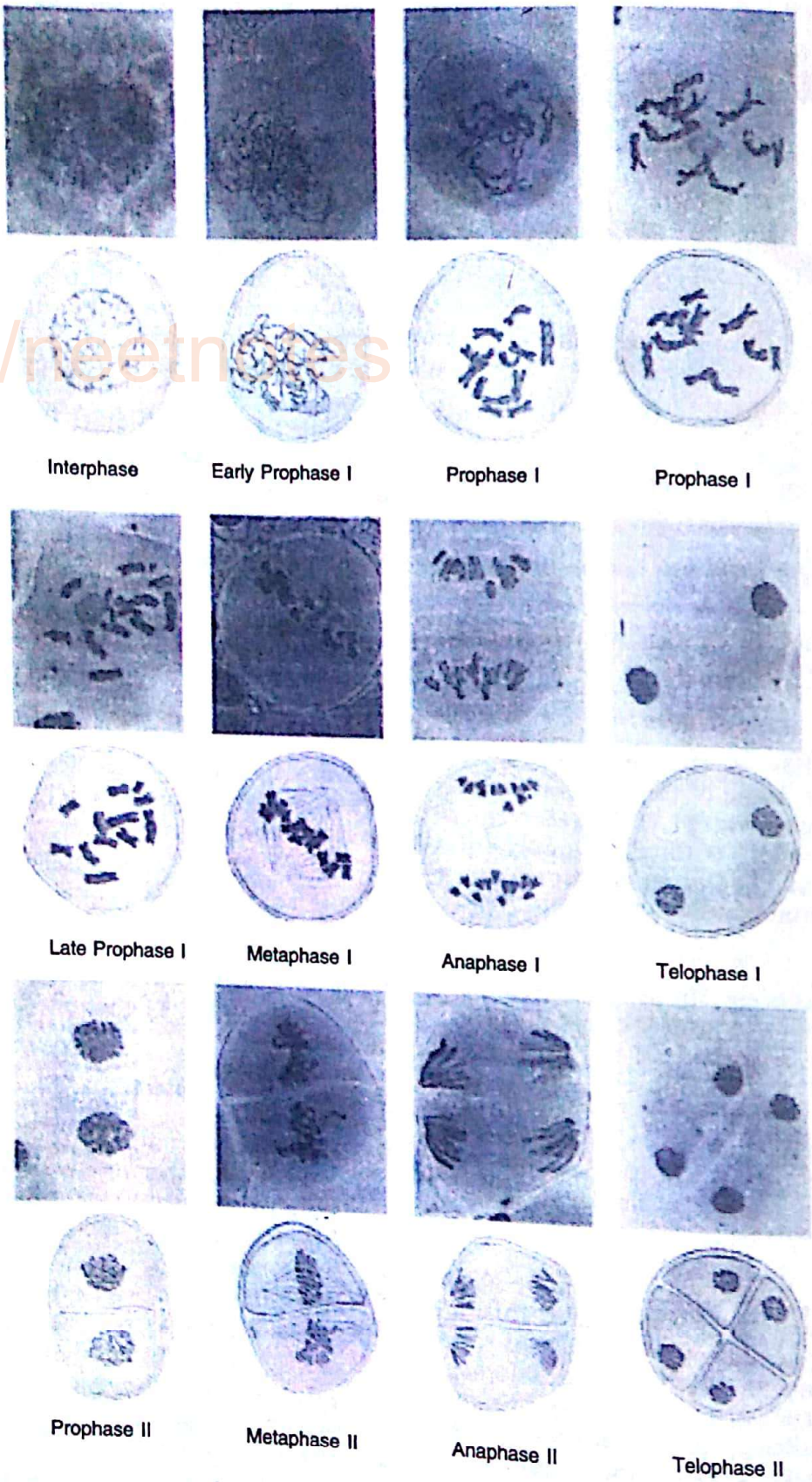

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Fig. 11.7 Stages of meiosis

(iii) Meiosis has genetic consequences due to events, such as
(a) pairing of the homologous chromosomes
(b) process of crossing over and recombination
(c) segregation of homologous chromosomes.

You can get a comparative idea of mitosis and meiosis from Table 11.1.

Table 11.1 Differences between Mitosis and Meiosis

Mitosis	Meiosis
The cell divides only once after one round of DNA replication.	There are two successive cell divisions, the first and the second meiotic divisions.
Mitosis takes place in the somatic cells.	Meiosis takes place in the germ cells.
It occurs in both sexually as well as asexually reproducing organisms.	It occurs only in sexually reproducing organisms.
The DNA replicates once for one cell division.	The DNA replicates once for two cell divisions.
The duration of prophase is short usually of a few hours.	Prophase I is comparatively longer and may take days.
Prophase is comparatively simple	Prophase I is complicated and is divided into leptotene, zygotene, pachytene, diplotene and diakinesis.
The cell divides only once and the chromosomes also divide only once.	There are two cell divisions but the chromosomes divide only once.
There is no synapsis.	Synapsis of homologous chromosomes takes place during prophase I.
The two chromatids of a chromosome do not exchange segments during prophase.	The chromatids of two homologous chromosomes exchange homologous segments during prophase at pachytene.
During prophase and metaphase each chromosome consists of two chromatids held together by a centromere.	During prophase and metaphase, homologous chromosomes form bivalents. Each bivalent has four chromatids and two centromeres.
The arms of the prophase chromatids are close to one another.	The arms of the chromatids are separated widely in prophase II.
Division of centromeres takes place during anaphase.	There is no centromeric division during anaphase I. Centromeres divide only during anaphase II.
Spindle fibres disappear completely in telophase.	Spindle fibres do not disappear completely during telophase I.
Nucleoli reappear at telophase.	Nucleoli do not reappear at telophase I.
The chromosome number remains constant at the end of mitosis.	The chromosomal number is reduced from the diploid to the haploid.
The genetic constitution of daughter cells is identical to that of parent cell.	The genetic constitution of the daughter cells usually differs from that of the parent cell due to crossing over. Each chromosome of daughter cells usually contains a mixture of maternal and paternal genes.

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SUMMARY

All eukaryotic organisms begin their life cycle as a single cell. Billions and trillions of cells are derived from this single cell by the process of cell division. There are two modes of cell division, mitosis and meiosis. The cell cycle represents a complete series of steps by which cellular material divides equally between daughter cells. The cell cycle has two periods: (i) Interphase – a period of preparation for cell division, and (ii) Mitosis the actual period of division. Interphase is further divided into G_1, S and G_2. In G_1 the cell grows. In the S phase the duplication of chromatin material occurs, i.e., synthesis of DNA, whereas during G_2, protein synthesis takes place. In animals, the centriole divides to form a new pair. During interphase, replication of chromosomes takes place, so that each chromosome now consists of two chromatids.

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Major stages in mitosis are prophase, metaphase, anaphase and telophase. Prophase is discernible by shortening and thickening of chromosomes. Simultaneously, the centrioles move to opposite poles, the nuclear envelope is dissolved, the spindle fibres start appearing and the nucleoli disappear. In Metaphase, the chromosomes are lined up in the centre to form equatorial plate. The centromere lies on the equator and arms towards the poles. During Anaphase, the centromere divides separating the two chromatids of each pair. The two sets of chromosomes migrate towards the opposite poles. Telophase begins when the two sets of chromosomes reach opposite poles. A new nuclear membrane is formed around each set of chromosomes. The nucleoli reappear. The chromosomes uncoil and lose stability. Cytokinesis refers to the division of cytoplasm. In an animal cell, a furrow appears between two daughter nuclei, it deepens and forms two daughter cells. In plant cells, a cell plate grows from middle to the periphery separating two daughter nuclei. All the cell organelles are distributed equally in both the daughter cells. Mitosis maintains the same chromosome number in daughter cells as the parent cell. It also restores surface; volume ratio of the cell. This division restores the nucleo cytoplasmic index. The growth in the body of organisms and repair of worn out parts takes place due to mitosis.

At prophase the nuclear envelope breaks down. The spindle is made up of microtubular protein and helps in anaphasic movement. As the chromosomes shorten, they also move.

Meiosis occurs in the reproductive cells and reduces the chromosome number to half in the daughter cells as compared to that of the parent cell. The meiotic cell division is divided into first and second meiotic phases – Meiosis I and II. In the first meiotic division, the homologous chromosomes pair to form bivalents, and exchange genetic material (chiasma) and then separate and get distributed into daughter cells. It has a long Prophase I divided into 5 substages: Leptotene, Zygotene, Pachytene, Diplotene and Diakinesis. During Metaphase I, the bivalents arrange on equatorial plate with their arms on the plate and centromere directed towards opposite pole. This is followed by Anaphase I in which the homologous chromosomes repel each other and go to the opposite pole with both their chromatids. Thus each pole receives half the chromosome number of the parent cell. In Telophase I, the nuclear membrane and nucleolus reappear. Meiosis II is similar to mitosis and the centromere of each chromosome breaks, separating the chromotids, one each to a daughter cell. The meiosis maintains chromosome number of a species. The four daughter cells are formed with half the chromosome number.

UNIT
FOUR

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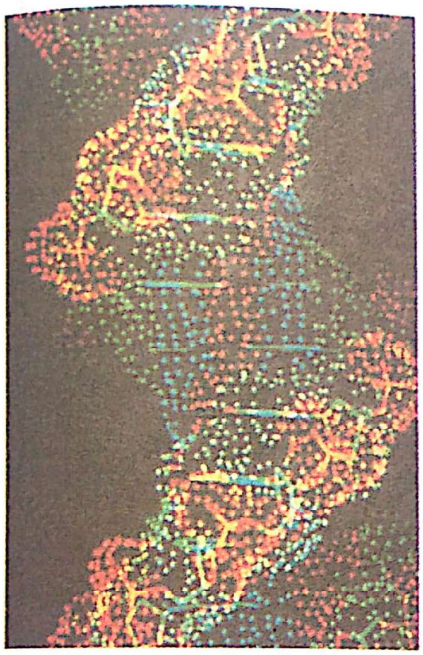
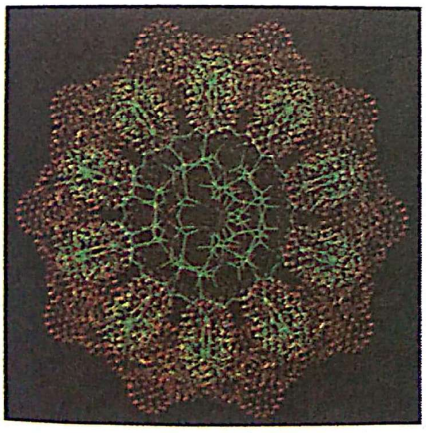
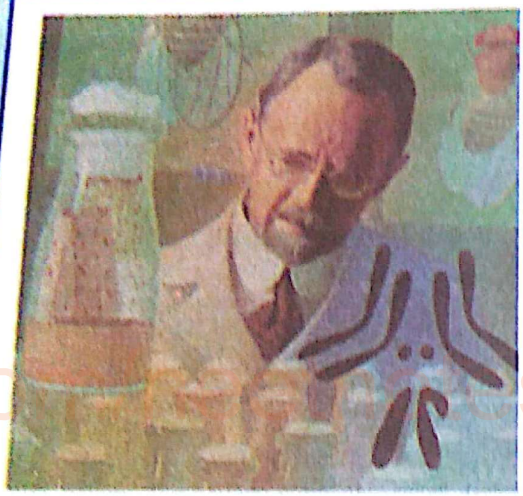




Genetics

Continuity of life is made possible by asexual and sexual reproduction. Sexual reproduction, besides creating new individuals, introduces variability in the offspring by combining traits of parents. How these traits are inherited and the laws that govern the inheritance remained a puzzle till they were unravelled by Mendel and others a little over a century ago. Now we know the units of heredity are genes that are transmitted from generation to generation. The parallel behaviour of genes and chromosomes during inheritance demonstrates the location of genes on chromosomes. The genes are arranged in a linear manner at specific positions on specific chromosomes. The molecule that is the carrier of information and programme of an organism's biological potential is deoxyribonucleic acid or DNA. In some viruses this function is performed by ribonucleic acid or RNA. Experiments have revealed the mechanisms how genes function in prokaryotic and eukaryotic cells. Further, it is beginning to reveal how different mechanisms regulate the expression of genes. But it is not fully understood why only a portion of coded information of DNA is expressed all the time whereas some others are expressed only when needed. Differential gene expression forms the molecular basis of normal processes of growth and differentiation as well as cancer. Advances in molecular biology have given us powerful tools and techniques to isolate individual genes and to transfer them from one kind of organism to another. These gene technologies are revolutionising agriculture and medicine. As scientists map genome, the total genetic content of organisms including our own, we look to the prospect of curing diseases, solving mysteries of forensics and biological evolution. This Unit will acquaint you with various aspects of genetics.



Thomas Hunt Morgan

(1866-1945)

American geneticist Thomas Hunt Morgan studied at Johns Hopkins University. Morgan's interest turned from embryology to the mechanisms involved in heredity. Morgan found that the rapidly multiplying Drosophila, the fruit fly, for studying how specific traits are transmitted through generations. Charting the family trees of fruit flies with such mutations as stunted wings, asymmetric bodies, and mismatched eye colouring, following the rediscovery of Austrian Scientist Gregor Mendel's work, Morgan elaborated the details of inheritance. He realised that there were more genes than chromosomes in Drosophila. He also discovered sex chromosomes and invented the techniques of genetic mapping. Thanks largely to Morgan's book, ***The Theory of the Gene*** *(1926), genetics was accepted as a legitimate branch of biology. He was awarded Nobel Prize for Physiology and Medicine in 1933.*

CHAPTER 12

GENETIC BASIS OF INHERITANCE

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In the previous units you have studied several aspects of organic life. Now we shall study how characters and traits are inherited from one generation to another. The different life forms may have similar patterns at the level of cellular organisation. A lot of diversity can be observed at the organismal level. While appreciating this diversity, one thing that must have struck you is the close resemblance between the members of a species. These resemblance are even stronger if we look at the descendants of a common ancestor, say parents and their progeny. Thus, every living organism possesses a set of characters by which it can be identified as a member of a particular species. What makes a species possess certain set of characters and how are they maintained generation after generation forms the subject matter of **genetics**. Although genetics is a relatively young discipline, it has gained immense importance in human affairs.

12.1 INHERITANCE : HEREDITY AND VARIATION

A great variety exists between the life forms. But if we observe closely they can be grouped on the basis of certain similarities into distinct types. For example, human babies are like human beings in overall characteristics, and a bitch will always produce puppies, which will have characters of its breed. This has been best summed up in the phrase 'like begets like'. If this phrase is analysed in the context of the knowledge of genetics, it would simply mean that there appears to be a continuity of life. In other words, this capability of maintaining a set of characters is passed on from one generation to another, a phenomenon known as heredity or inheritance.

Also implicit in this similarity are the differences or the variations within a species. After all, we can easily distinguish the members of a family, except the identical twins. Similarly, we can differentiate our own pet dog from other dogs. So, hereditary component must also explain some of these variations that exist within a species. As we shall see later, heredity (the similarity of offspring to parents) and variations (the differences between parents and offsprings, and among themselves) are the two aspects of the same fundamental mechanism.

Another component of heredity and variation is associated with the mode of reproduction of a species. The variation in sexually reproducing organisms, such as most animals, plants and some microbes, can be easily observed. Many lower organisms like microbes, fungi, some plants, and some lower animals, reproduce asexually. Here the progenies are derived from a single parent and thus receive the characters from that parent alone.

12.2 PRE-MENDELIAN IDEAS ABOUT INHERITANCE

It is difficult to say when people first conceived the idea of heredity. A variety of archaeological evidence, such as cave paintings and stone carvings document successful domestication of animals and cultivation of plants thousands of years ago. Thus, ancient man learnt both heredity and artificial selection of genetic variations. One idea that persisted for a long time was that the characteristics from the parents some how 'blended' at conception and offsprings were essentially a 'dilution' of the different parental characteristics.

Charles Darwin who is known for his theory of natural selection, also proposed the hypothesis of pangenesis. This hypothesis suggested that every part of the body produces a representative 'gemmule' which, by the blood circulation, gets collected in the semen. These 'gemmules' then transmit the characteristics to the offspring. In 1892, August Weismann discounted the concept of gemmules and proposed that living organisms consist of two kinds of materials–somatoplasm and germplasm. The somatoplasm makes the whole body that undergoes growth, development and ultimately death. He believed that the germplasm is immortal and provides continuity among succeeding generations.

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12.3 GREGOR JOHANN MENDEL AND HIS THEORY OF INHERITANCE

Let us see how Gregor Mendel discovered his Theory of Inheritance. In 1856, Mendel started his experiments on controlled hybridization in garden pea in the monastery garden at Bruno. In 1865, he first reported the results of some of his experiments but these went unnoticed and unappreciated until 1900, when the work was rediscovered independently by three geneticists. Mendel is, thus, appropriately called the 'Father of Genetics'.

Mendel's Experiment

Mendel chose garden pea (*Pisum sativum*) for his experiments. But why did he choose this plant? Because the pea plant is small, easy to grow and cross bred artificially. This plant, being bisexual, is self-fertilising in nature but can be easily cross-pollinated experimentally. It reproduces a large number of offspring and completes its life cycle in one season. A large number of true breeding varieties of peas are available. Mendel selected seven visible *characters*, each with two contrasting *traits* (Fig.12.1). He also kept accurate records of his experiments, giving all the details of number and type of individuals, which are a necessity in the genetic studies.

Experimental Method

Mendel's experiments were based on cross-breeding two plants of pea differing with each

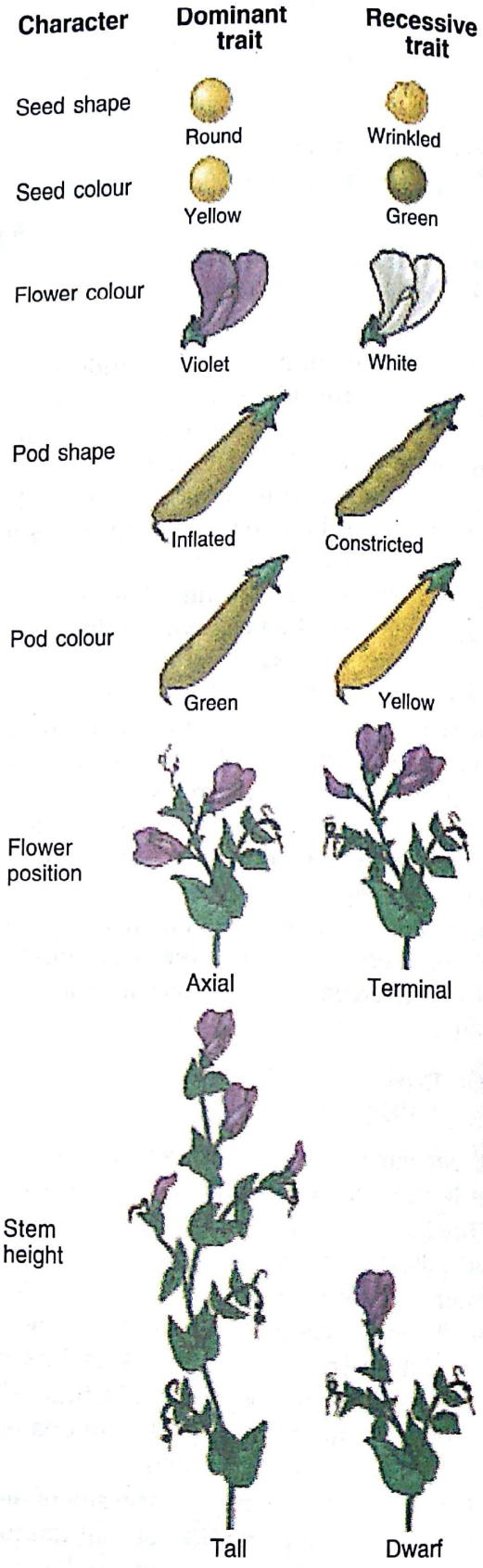

Fig 12.1 *Seven pairs of contrasting traits in pea plant*

other in certain contrasting traits, which are listed in Table 12.1.

The simplest of the crosses conducted by Mendel, called **monohybrid** cross consisted of plants differing in one of the contrasting pair of

Table 12.1 Seven Pairs of Contrasting Traits in Pea used by Mendel for his Experiments

S.No.	Characters	Contrasting traits
1	Stem height	Tall/dwarf
2	Flower colour	Violet/white
3	Flower position	Axial/terminal
4	Pod shape	Inflated/constricted
5	Pod colour	Green/yellow
6	Seed shape	Round/wrinkled
7	Seed colour	Yellow/green

traits, for example, tall or dwarf, violet- flowered or white-flowered, etc. A cross was made by taking the pollen grains from one type of plant to fertilise the egg of the other (mating or hybridization) (Fig.12.2), and then collecting the offspring in the form of seeds. Mendel extended his work by including two or three pairs of contrasting traits in his crosses which were called **dihybrid** and **trihybrid** crosses. The process of crossing remained the same as described above except that the two parents differed from each other in one, two, or three of the seven characters. The first generation offspring (F_1) were selfed or self-fertilized to give rise to the second generation progeny (F_2).

Results of the Experiment

The original parents in a cross are called Parental or P generation, their offspring are the First Filial generation or F_1 and the offspring derived from selfing of the F_1 are termed as Second Filial or F_2 generation. Let us now examine Mendel's data cross-wise and try to understand the principle of inheritance as proposed by him.

A cross between tall-stem versus dwarf-stem plants is a typical monohybrid cross. When true-breeding tall plants were crossed with true-breeding dwarf plants, the F_1 generation resulted in all tall plants. When these F_1 talls

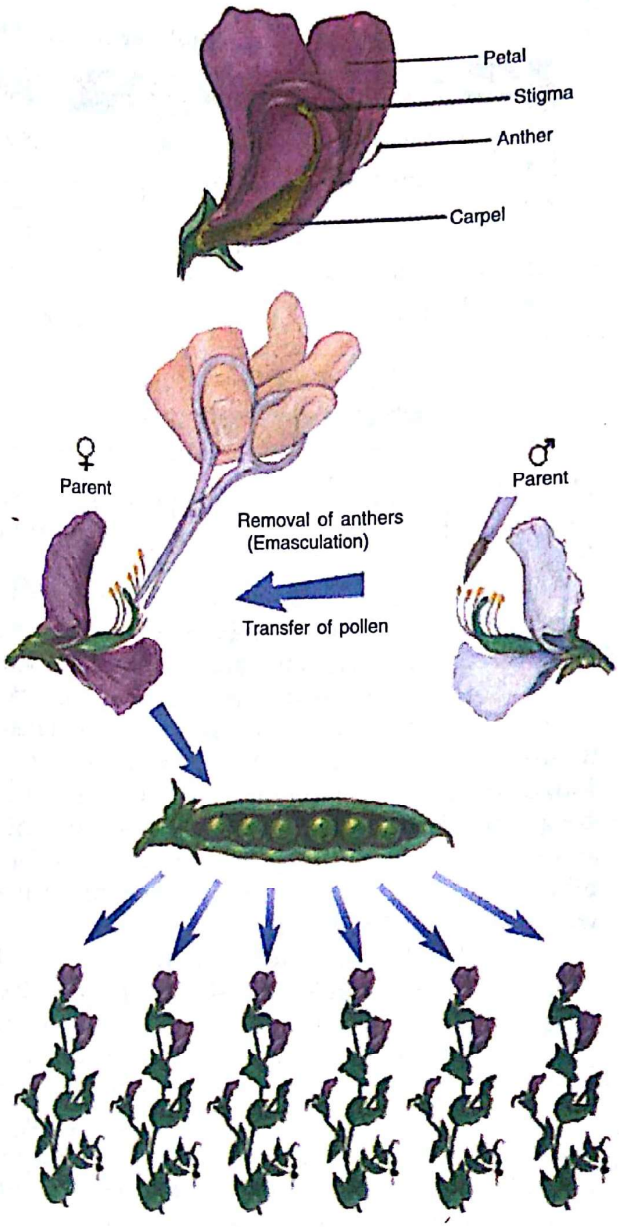

Fig 12.2 *Steps in making a cross in pea*

were selfed, however, Mendel observed that out of the total 1064 F_2 progeny, 787 were tall and 277 were dwarf. You must note here that dwarfness which disappeared in F_1 generation reappeared in F_2. These data are not derived from a single cross but represent the results of several tall x dwarf crosses, F_1 selfing and the analysis of total 1064 F_2 offspring. In order to put these results in a more rational way, the absolute numbers were converted to ratios and this has become a normal genetic convention.

Table 12.2 Analysis of Monohybrid Crosses

S.No.	Cross	F_1	F_2	Ratio
1.	Tall × dwarf stem	Tall stem	787 tall, 277 dwarf	2.84:1
2.	Violet × white flower	Violet flower	705 violet, 224 white	3.15:1
3.	Axial × terminal pod	Axial pod	651 axial, 207 terminal	3.14:1
4.	Full × constricted pod	Full pod	882 full, 299 constricted	2.95:1
5.	Green × yellow pod	Green pod	428 green, 152 yellow	2.82:1
6.	Round × wrinkled seed	Round	5474round,1850 wrinkled	2.96:1
7.	Yellow × green seed	Yellow	6022yellow, 2001green	3.01:1

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Mendel thus found that the offspring appeared in a ratio of 2.84 tall : 1 dwarf or approximately 3:1.

He made similar crosses with other pairs of contrasting traits and in every case the outcome was the same. In each, F_1 plants were identical to one of the parents and the F_2 generation was divided in the approximate ratio of 3:1. Once again, three-fourth were like F_1 parent and one-fourth expressed the contrasting trait. It must be pointed out that in each cross a large population of F_2 progeny was analysed and in all cases the approximate ratio of a pair of traits was 3:1 (Table 12.2).

It should also be noted that the pattern of inheritance in F_1 and F_2 was similar irrespective of which plant served as the female and which served as the male. In other words, the outcome of a monohybrid cross is just the same whether violet-flowered plant pollinated the white-flowered female plant or vice-versa. Such crosses are called **reciprocal crosses**. The consistent pattern of results obtained from different monohybrid crosses led Mendel to propose four postulates which have been called **Principles of Inheritance**.

Postulate 1

Mendel proposed that each genetic character is controlled by **a pair of unit factors**, now commonly called **alleles or allelomorphic pair**. If we analyse a monohybrid cross in the light of this postulate in the true-bred parents, there are two unit factors for tallness and similarly two unit factors for dwarfness. Thus, three combinations are possible: either there are two factors for tall or two factors for dwarf stems, or one of each factor in an individual.

Postulate 2

Based on the results obtained in F_1 generation, Mendel was able to propose that when two dissimilar unit factors are present in a single individual, only one is able to express and the other is not. The one that expresses itself is the **dominant unit factor** and the other which fails to express is the **recessive unit factor**. These terms are also used to denote a trait, for example, tall stem is said to be dominant over the recessive trait dwarf stem. We now call these unit factors **"genes"**.

Postulate 3

During gamete formation , the unit factors of a pair separate or segregate randomly so that each gamete receives one or the other unit factor with equal probability. This ensures the purity of gametes. With this postulate, we can extend our analysis up to F_2 generation of the crosses shown in Table 12.2. When gametes are formed in P generation (tall x dwarf) each gamete from the parent will receive one but the same unit factor either for tallness or dwarfness. Following fertilisation, F_1 plants will receive one unit factor for tallness and one for dwarfness. With the former being dominant, all F_1 plants will be tall. When F_1 tall plants will form gametes, the principle of segregation will demand that each gamete randomly received either the tall or the dwarf unit factor. With subsequent fertilisation, which is a chance event, four combinations of these unit factors will result in equal frequency in F_2 generation: (i) tall/tall, (ii) tall/dwarf, (iii) dwarf/tall, (iv) dwarf/dwarf (Fig. 12.3).

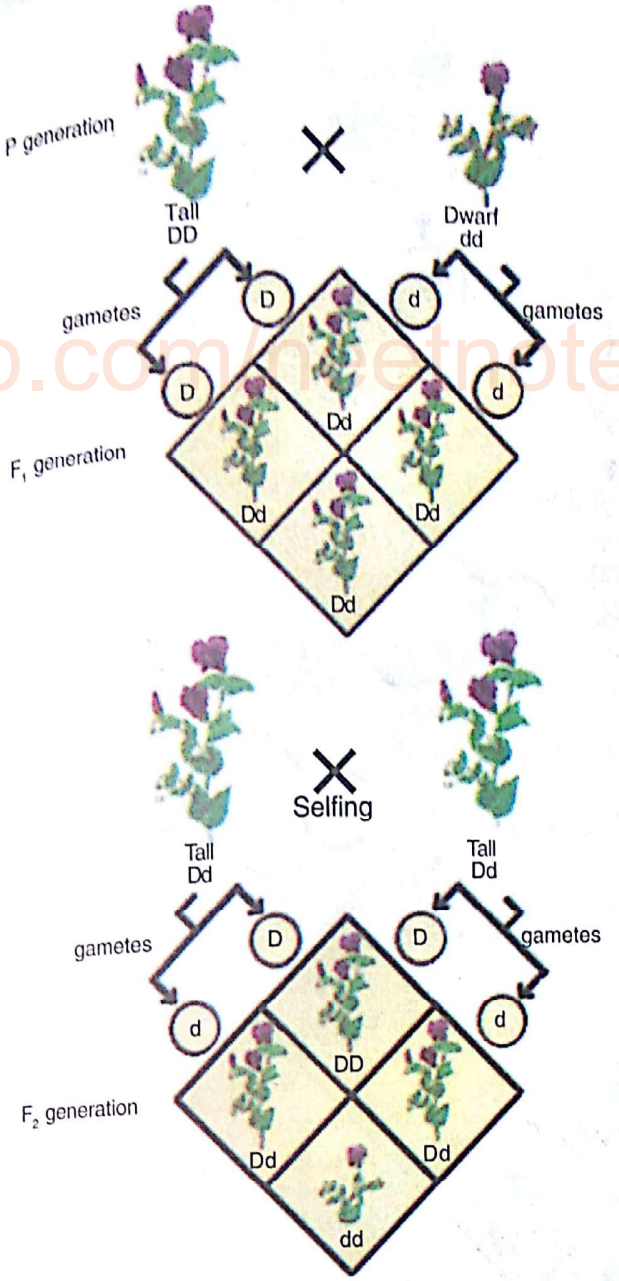


Fig 12.3 *A typical monohybrid cross conducted by Mendel between true-breeding tall plants and true-breeding dwarf plants*

If we apply the postulate 2, the combinations (i), (ii), and (iii) will result into tall plants, and only combination (iv) will give the dwarf ones. In other words, the F_2 generation is predicted to consist of three-fourths tall and one-fourth dwarf or a phenotypic ratio of 3:1. This is exactly what Mendel had observed, and is seen in all the monohybrid crosses. If we apply the postulate 3, the combinations (i) and (iv) will result into homozygous tall and homozygous dwarf respectively, and combinations (ii) and (iii) will result into heterozygous tall, and the genotypic ratio will be 1:2:1 for homozygous tall, heterozygous tall and homozygous dwarf, respectively in F_2 generation. The postulate 3 is popularly known as the **Principle of Segregation** or **Purity of Gametes**.

When any individual produces gametes, the **alleles segregate** or separate; so each gamete receives only one member of the pair of alleles (and the paired condition is restored by random fusion of gametes during fertilisation).

Postulate 4

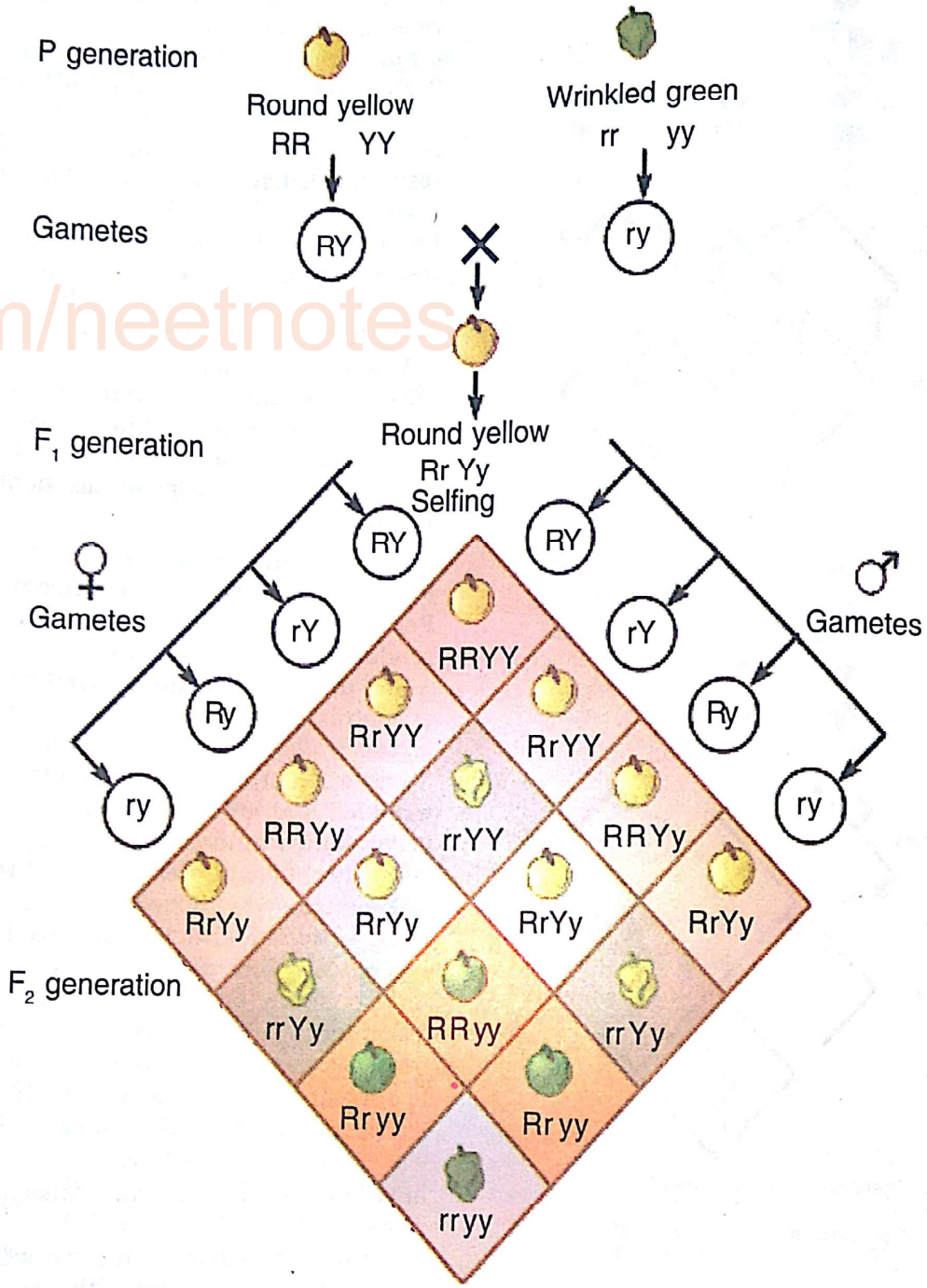
This postulate is also referred to as the **Principle of Independent Assortment of Factors**. Mendel extended his work by conducting dihybrid or two factor crosses, in which the parent plants differed in two pairs of contrasting traits. Let us consider a cross in which plants producing round and yellow seeds were crossed with plants producing wrinkled and green seeds. The F_1s were all yellow and round-seed ed; suggesting that yellow is dominant over green and round seed is dominant over wrinkled seed. When these F_1s were selfed, Mendel postulated that the segregation of one pair of unit factors will occur independently of the other pair or they will assort independently. Accordingly, the gametes must carry all possible combinations of the unit factors in equal frequency. In this example, gametes with random distribution of unit factors will give following combinations:

(i) yellow, round; (ii) green, wrinkled; (iii) green, round; and (iv) yellow, wrinkled.

During fertilisation, each zygote will have equal probability of receiving two of these combinations, one from each parent. This will result into a progeny which can be classified as

9/16 yellow, round;
3/16 yellow, wrinkled;
3 /16 green, round; and
1/16 green, wrinkled.

This prediction turned out to be true with Mendel's dihybrid data (Fig. 12.4).



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Phenotypic ratio : round yellow : round green : wrinkled yellow : wrinkled green
9 3 3 1

Genotypic : RRYY : RrYY : RRYy : rrYY : RrYy : rrYy : RRyy : Rryy : rryy
1 : 2 : 2 : 1 : 4 : 2 : 1 : 2 : 1

Fig 12.4 *Results of a dihybrid cross where the two parents differed in two pairs of contrasting traits such as seed colour and seed shape*

If the two pairs of contrasting traits are inherited independently, the principle of independent assortment can also be stated as *when two independent events occur simultaneously, the combined probability of two outcomes is equal to the product of their individual probabilities of occurrence.* For example, the segregation of yellow x green and so also of
round seed x wrinkled seed is 3/4 yellow: 1/4 green, and 3/4 round seed: 1/4 wrinkled seed.

If we combine the two events:
(3/4) yellow x (3/4) round seed
= 9/16 yellow, round;
(3/4) yellow x (1/4) wrinkled seeds
= 3/16 yellow, wrinkled;
(1/4) green x (3/4) round
= 3/16 green, round seeds; and
(1/4) green x (1/4) wrinkled
= 1/16 green, wrinkled seeds.
This is the ratio obtained in a dihybrid cross (Fig. 12.4).

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12.4. GENETIC TERMS AND SYMBOLS

In order to develop a better understanding of Mendel's work and other genetic phenomena, we must familiarize ourselves with some terms and terminologies routinely used in genetic studies (Table 12.3).

Table 12.3 Genetic Terminology

Terms	Meaning	Examples
Character	It is the feature of the individual.	Stem height
Trait	An inherited character and its detectable variant.	Tall or dwarf
Unit factor	A unit of inheritance called gene by modern geneticists. Each gene or factor controls a character.	D and d
Allele	It represents atleast two alternative forms of a gene or unit factor. Each gene consists of an allelomorphic pair.	Dd, Dd, Dd
Gene symbol	Each trait is provided a symbol. By one convention the first letter of the mutant trait is chosen to derive the symbol, e.g. dd for dwarfness and DD for tallness (this is sometimes referred as tt and TT, respectively).	
Phenotype	It is observable morphological appearance. The phenotypes of an individual is determined by different combinations of alleles.	Tall or dwarf
Genotype	It is representation of an individual's genetic constitution with respect to a single character or a set of characters.	DD Dd dd
Homozygous	When the two alleles of a gene are similar and so two copies of the same allele exist, they are said to be in homozygous combination.	DD or dd
Heterozygous	When the two alleles in a pair are different they are in heterozygous state.	Dd
Dominant	An allele that influences the appearance of the phenotype even in the presence of an alternative allele.	D
Recessive	An allele that influences the appearance of the phenotype only in the presence of another identical allele	d

One can seek the answer to the question as to why did Mendel propose pairs of unit factors for each character, more meaningfully now. Firstly, each character had two contrasting forms (traits) which logically should be governed by two distinct unit factors. Secondly, though the F_1 showed one phenotype (that of the dominant trait), the unit factor for recessive trait did not disappear or blend. It was simply masked or hidden and reappeared in the F_2 progeny on selfing the F_1.

Mendel's analytical reasoning must be considered truly amazing. He was able to propose that heredity is governed by discrete particulate units on the basis of some simple but precisely executed breeding experiments. He thus explained how traits are transmitted from one generation to the next.

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12.5 METHODS OF ANALYSIS

Punnett Square

The genotypes and phenotypes resulting from various combinations of gametes can be easily determined by Punnett squares, devised by Reginald.C. Punnett (1875-1967). Here, each of the possible gamete is placed in an individual column or a row, with vertical column representing the female and horizontal row the male parent. The gametes are then arranged in all possible combinations and the resulting genotypes are entered in the boxes along with phenotypes (Figs. 12.3, 12.4).

Test Cross

You have learnt so far, that Mendelian crosses involved two pure-breeding parents which shall yield a heterozygous F_1. In order to prove that F_1 is heterozygous, Mendel devised a simple cross called **test cross**. In a test cross, the F_1 generation is crossed to a recessive parent instead of selfing. The results can be easily analysed (Fig. 12.5). If you follow the monohybrid cross where the F_1 is test crossed, a ratio of 1:1 will be obtained. On the same basis, you can work out that in a dihybrid case, the test cross ratio will be 1:1:1:1. Test cross can also be used for another purpose. You must have understood by now that the homo- and heterozygous genotypes for dominant trait cannot be differentiated because they show the same phenotype. If we put them through a test cross, you will see that all homozygous dominant combinations will breed true but heterozygous genotypes will follow the segregation.

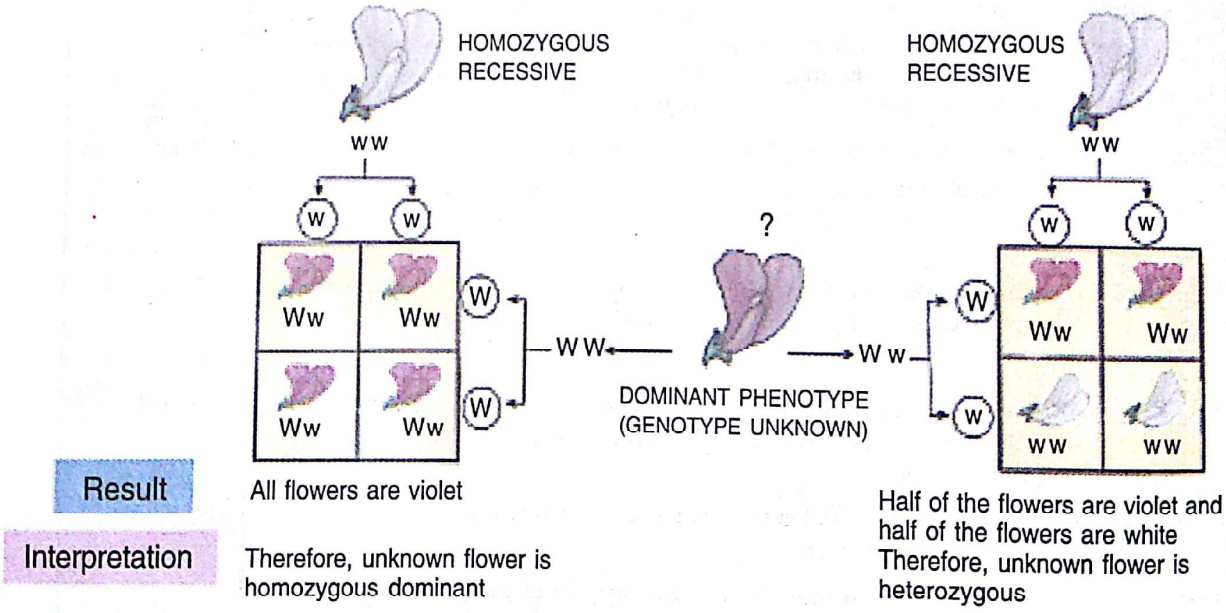


Fig 12.5 *A test cross can be conducted to differentiate between a homozygous and a heterozygous dominant genotype*

Trihybrid Cross

After analysing dihybrid crosses, Mendel also conducted some trihybrid or three-factor crosses. The results of such crosses are some what complicated but can be easily analysed by applying the principles of segregation and independent assortment.

12.6 REDISCOVERY OF MENDEL'S WORK

Mendel started his work on pea in 1856 but presented it for the first time in 1865. The work was published in the following year, but remained unrecognised for about 35 years due to several reasons. Firstly, Mendel's ideas on heredity appeared to be too simple in contrast to the views held by his predecessors. Also, Mendel's mathematical approach to solve a biological phenomenon was completely alien for his contemporaries. Moreover, Mendel by postulating discrete or particulate units referred to what came to be known as discontinuous variation, whereas his strong contemporaries like Charles Darwin and Alfred Russell Wallace advocated the significance of continuous variation in evolution. With the whole emphasis being concentrated on organic evolution at this time, Mendel's work failed to create the necessary impact on the scientific community.

In the early twentieth century, hybridization experiments similar to Mendel were performed independently by three botanists, Hugo deVries, Carl Correns and Erich Tschermak-Seysenegg. This led to the rediscovery of the Mendelian work and the birth of the Science of Genetics.

12.7 POST-MENDELIAN ERA - OTHER PATTERNS OF INHERITANCE

After the re-establishment of Mendel's laws of inheritance in 1900, one common approach employed by geneticists was to repeat Mendel's experiments using other organisms and different phenotypes. Although on the surface, it may look repetitive, this period provided a flush of information on how genes control the phenotype. It became clear that a phenotype may be an outcome of different interactions rather than a mere one to one relationship. Such interactions may occur at allelic as well as non-allelic levels and provide other patterns of inheritance.

Before we study these patterns, let us look at other genetic conventions of writing gene symbols. As we discussed earlier, an allele is

GREGOR JOHANN MENDEL
(1822-1884)

In 1822, in a small village of Heinzendorf in Austria (now a part of Czech Republic), in a peasant family was born a boy named Johann Mendel, who was to make singular contributions in the field of Genetics. Mendel was a brilliant student in school and studied philosophy for several years before joining the Augustinian Monastery of St. Thomas in Bruno in 1843. There he took the name Gregor and received support for his studies and research throughout his life. In 1849, he took up a teaching appointment. Later, he joined the University of Vienna from 1851-1853 to study Physics and Botany and returned to Brno to teach physics and natural sciences.

In 1856, Mendel performed his first set of hybridization experiments with the garden pea and followed it up very meticulously for the next ten years or so. His interest in genetics remained alive even when he did not get any recognition for his work. He died in 1884 of a kidney disorder.

identified by its mutant phenotype, and symbol (consisting of initial letter or preferably three letters) is derived from the same. If recessive, a lower case letter and if dominant an uppercase letter is used to represent the symbol. Since the mutant allele is derived from a wild-type allele, the latter carries the same symbol but with a superscript $^+$.

Incomplete Dominance

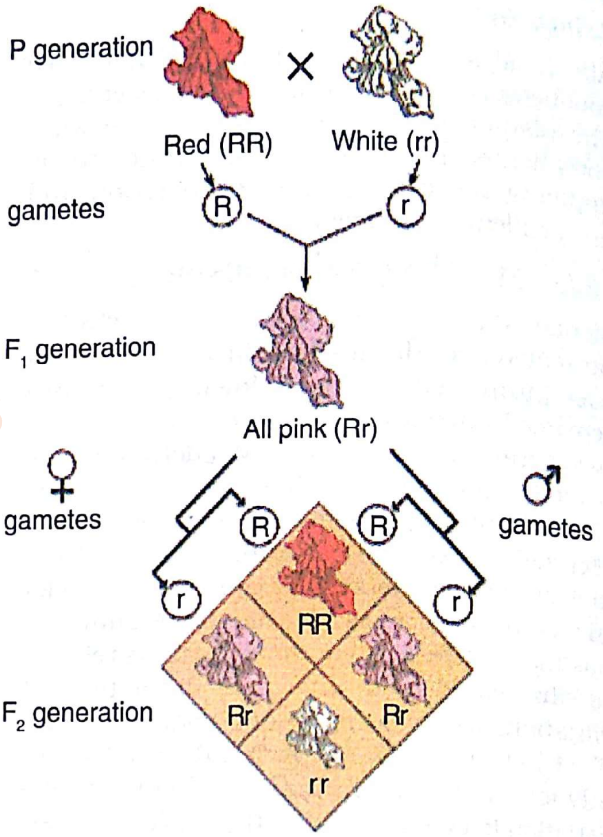
We have learnt so far that between two alleles of a gene, one is completely dominant over the other and, therefore, does not allow the expression of the recessive allele in a heterozygote. In plants like four-O-'clock (*Mirabilis jalapa*) and Snapdragon (*Antirrhinum majus*), if red-flowered plants are crossed with white-flowered ones, the F_1 produces pink flowers. Dominance thus appears to be partial or incomplete. That this is not a case of blending inheritance, as one might tend to believe (unlike Mendel), can be understood if we look at the results of F_1 (pink) x F_1 (pink) cross (Fig. 12.6).

In F_2, a genotypic ratio of 1:2:1 is obtained like a typical monohybrid cross with the difference that phenotypic ratio is also 1:2:1. The clear-cut cases of incomplete dominance at phenotype level may be rare. In a heterozygote, the recessive allele may also express resulting into a different phenotype.

Other Allelic Interactions

Besides incomplete dominance, alleles could also show **codominance**. Here in a heterozygote both alleles express themselves. This phenomenon has been observed in MN blood groups in humans. The red blood cells can carry two types of native antigens, M and N, and an individual can be MM, MN, or NN, exhibiting either one or both of them.

Similarly, the products of some genes can be essential for the survival of an organism. Mutation in this gene that leads to non-functional product will affect the viability. These are called lethal mutations; and two types can be encountered. If one dominant allele is sufficient for the normal phenotype, the heterozygote will survive (recessive lethal allele). However, if the heterozygote does not survive then mutation has created a dominant lethal allele.



Phenotypic ratio : red : pink : white
1 : 2 : 1

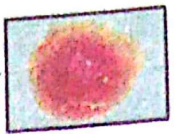
Genotypic ratio : RR : Rr : rr
1 : 2 : 1

Fig 12.6 *Results of monohybrid cross in the plant Snapdragon, where one allele is incompletely dominant over the other allele*

Multiple Alleles

We have so far learnt about two alternative forms of a gene and thus two alleles governing the same character. From the genetic studies in the post-Mendelian era, it started becoming clear that three or more alleles of a gene can be found. The mode of inheritance in such cases was called

Blood type (Phenotype)	Genotype	Antigen	Antibodies	Reaction to added antibodies	
				Antibody-a	Antibody-b
A	I^AI^A or I^AI^O	A	b		
B	I^BI^B or I^BI^O	B	a		
AB	I^AI^B	Both A and B	Neither a nor b		
O	I^OI^O	Neither A or B	Both a and b		



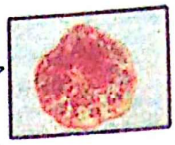


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Fig 12.7 *Detection of A, B and O blood type in humans determined by multiple alleles and two alleles acting codominantly over third*

multiple allelism. In any diploid individual, only two alleles can be found, so multiple alleles can be detected only in a population.

The simplest example of multiple alleles is the inheritance of A,B and O blood groups in humans. In human populations, three different alleles for this character can be found: I^A, I^B, and I^o. While I^A and I^B are responsible for A and B antigens, respectively, I^o does not produce any detectable antigens. The blood typing in humans, therefore, can be determined by their antigen types, as shown in Fig. 12.7.

Human blood typing has several practical applications. Firstly, the compatibility of blood types can be matched during blood transfusion and secondly, the cases of disputed parentage can be settled more accurately.

Several other phenotypes in plants and animals are governed by multiple alleles. The eye colour gene in *Drosophila* and self-incompatibility genes in some plants may have large number of alleles.

12.8 TWO GENE INTERACTIONS

Better understanding of gene expression at the level of phenotype also led to the examples where a phenotype is not controlled by one gene but is an outcome of the contribution of more than one non-allelic genes. Such genes are bound to show an interaction at the phenotypic level depending upon the type of the alleles carried by an individual. We shall discuss below some cases of two gene interactions, for which we shall make some assumptions and use some conventions:

(i) In each case, a distinct phenotype is produced.

(ii) Genes under consideration show normal dominance and assort independently.

(iii) On phenotypic level, homozygotes and heterozygotes, such as AA or Aa and BB or Bb, will show their respective phenotype governed by the dominant allele. They can thus be designated as A– or B–. In other words, a dash indicates that either allele may be present.

(iv) The F_1 must be heterozygous for the two gene pairs (Aa Bb) which will be selfed or intercrossed.

(v) In a normal dihybrid cross, the F_2 genotypes fall into four phenotypic classes 9/16 A–B–, 3/16 A–bb, 3/16 aa B–, and 1/16 aabb.

Epistasis

The best examples of gene interaction are those illustrating the phenomenon of epistasis. Here, one gene masks or modifies the expression of another non-allelic gene. The gene which can override the expression of the other is the epistatic gene and the one which gets masked is the hypostatic gene. Either of the two alleles,

Table 12.4 Some Common Examples of Gene Interaction

S.No.	F_1 phenotype (AaBb x AaBb)	F_2 genotypes				Phenotypic ratio & explanation
	Example	9A–B–	3A–bb	3 aaB–	1 aabb	
1	Coat colour in Mouse	Agouti	Albino	Black	Albino	9:3:4 recessive epistasis (supplementary genes)
2	Fruit colour in Squash	White	White	Yellow	Green	12:3:1 dominant epistasis
3	Flower colour in Pea	Purple	White	White	White	9:7 complementary genes (both dominant alleles required for a phenotype)
4	Fruit shape in Squash	Disc	Circular	Circular	Long	9:6:1 new phenotype by either allele
5	Fruit shape in Shepherd's purse	Triangular	Triangular	Triangular	Ovoid	15:1 either allele separately or together give same phenotype
6	Feather colour in Fowls	White	White	Coloured	White	13:3 inhibitory gene (one gene inhibits the expression of the other)

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recessive or dominant, can bring about the epistatic effect. In some cases, some new phenotypes may be generated due to such interactions. All these will lead to a modified dihybrid ratio as shown in Table 12.4. It should be emphasised that while epistasis is the interaction between different genes (non-alleles), dominance is the interaction between different alleles of the same gene.

We shall now discuss one of these crosses involving coat colour in detail. You should be able to work out other examples based on the assumptions and explanations.

In this example, the coat colour is determined by A/a pair with A- giving agouti and aaB black phenotype. Of the B/b allele pair, recessive allele b is epistatic over A/a. Thus, in the presence of bb, both A- and aa give the same phenotype (albino). This will serve as an example of recessive epistasis (Fig. 12.8).

By going through these examples, you must keep in mind that we are not deviating from Mendel's Principles of Segregation and Independent Assortment. The differences that you see is only at the phenotypic level and that too because the phenotype is under the control of two genes.

12.9 POLYGENIC TRAITS

Mendel had described discrete differences between the traits that can alternatively be expressed as discontinuous variation. In other words, the phenotypic differences were distinct: tall or dwarf plant, violet or white flower, green or yellow seed, etc. But it is not always so. If you look around in your class itself, you can pick up two important examples of what is called continuous variation. These are human height and skin colour. You can observe that there are no distinct alternative forms, instead you have all the different grades of the phenotype. Several other characters such as seed size and seed colour, plant height and yield in plants are the other common examples. Such a trait is called **polygenic** or **quantitative**. Though polygenic traits can be easily influenced by environment, they are generally controlled by three or more genes with phenotype reflecting the contribution

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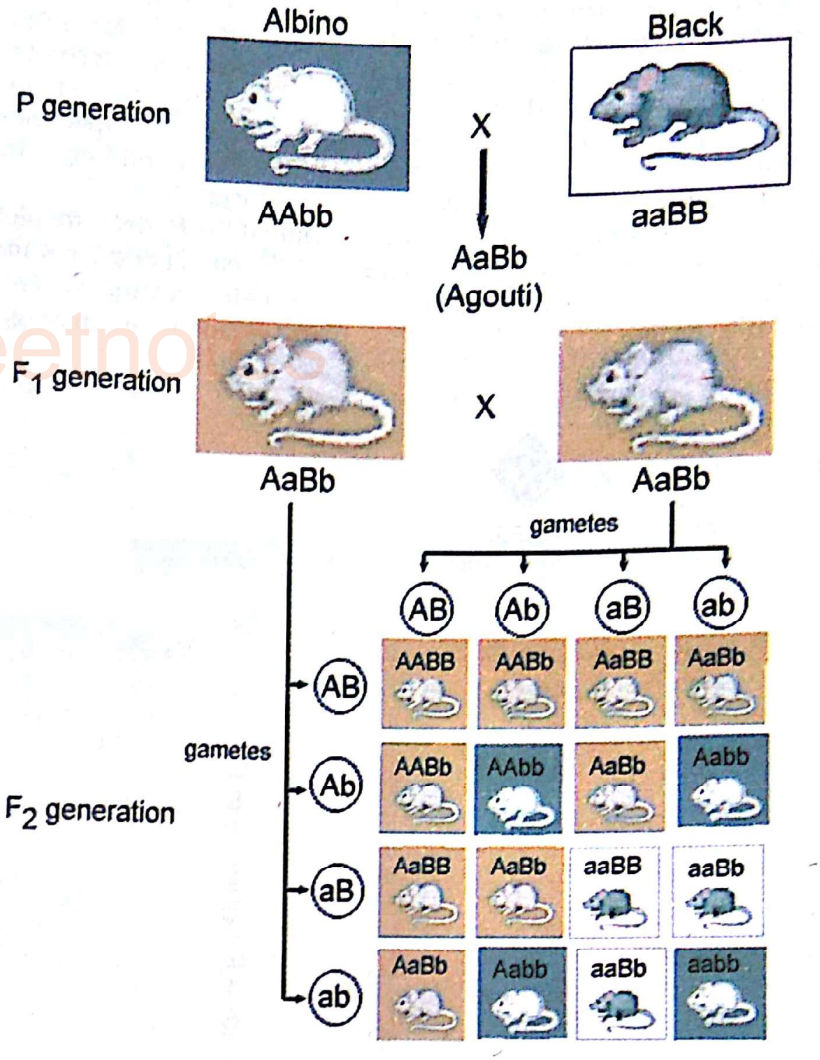

Phenotypic ratio : agouti : black : albino
9 : 3 : 4

Genotypic ratio : AABB : AABb : AaBB : AaBb : AAbb : Aabb : aaBB : aaBb : aabb
1 : 2 : 2 : 4 : 1 : 2 : 1 : 2 : 1

Fig 12.8 *Inheritance of a character determined by two genes in which one allele is epistatic over the other allele pair*

of each allele (quantitative). Let us discuss this polygenic or multiple gene trait by studying the inheritance of human skin colour. By now, it should be clear to you that there are no contrasting phenotypes for this trait. Let us also assume that this trait is controlled by three genes, A, B, and C. In the cross, we are following, there is a mating between dark-skinned and fair-skinned human beings, and then the intermediate skin coloured individuals expected at F_1 are mated (Fig 12.9a).

It is clear that

(i) fewer and fewer individuals fall into parental categories;

(ii) the expression level of the phenotype was dependent upon the number of contributing alleles and hence was more quantitative; and

(iii) if the F_2 data are plotted graphically, a bell-shaped curve results (Fig. 12.9b).

In this example, we have assumed the involvement of three gene pairs, however, if

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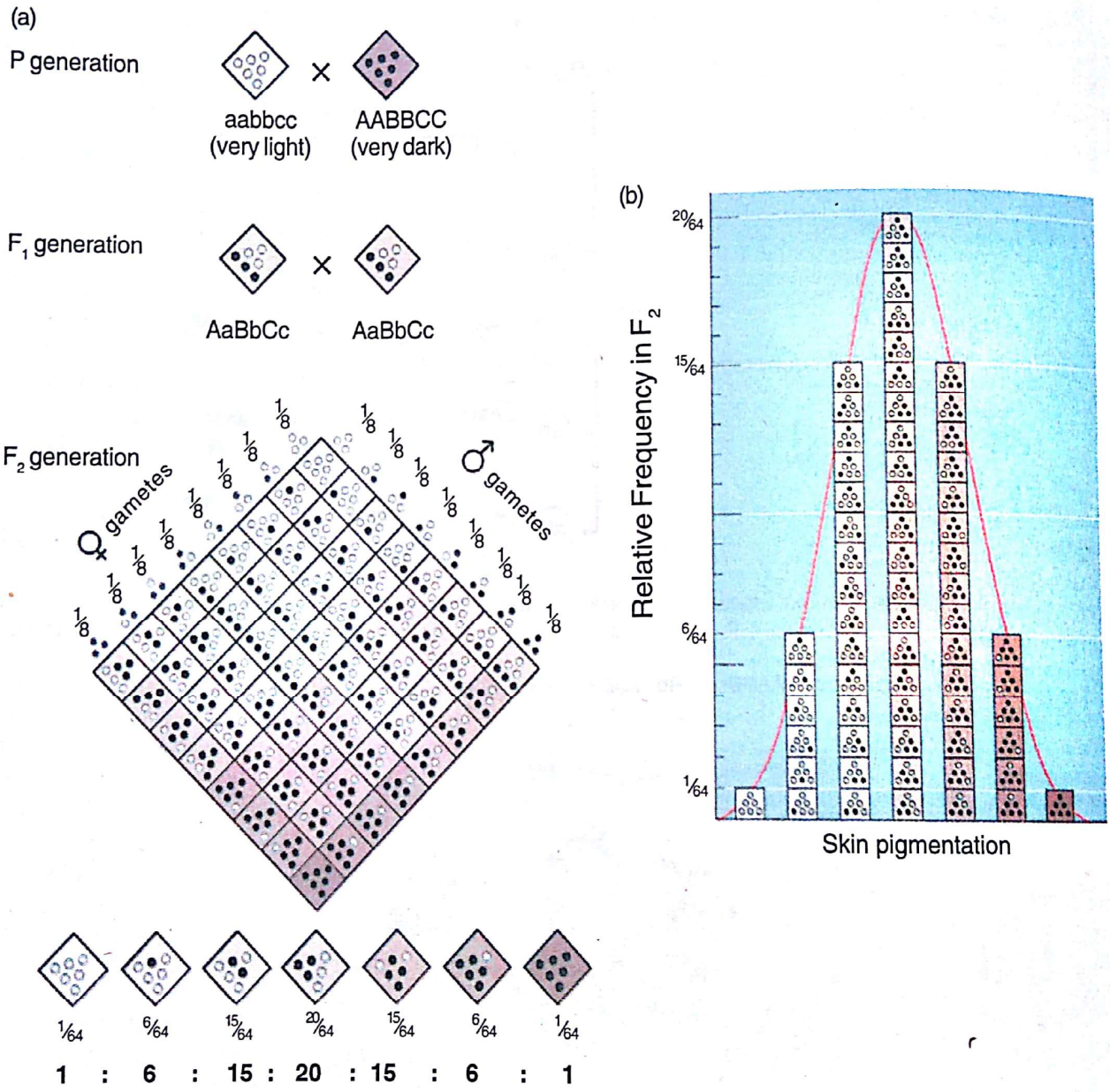

Fig 12.9 (a) A cross depicting the inheritance of human skin colour controlled by polygenes, (b) If the relative frequency of F_2 progeny in a polygenic cross is plotted against the extent of phenotypic expression a typical inverted bell shaped curve is seen

higher number of genes are involved in determining a phenotype, greater variety would be expected in the F_2 generation. By studying several examples from plants and animals, it became an accepted fact that a large number of characters are controlled by polygenes in which alleles contribute additively to a phenotype. The total contribution of each additive allele at each locus may be small but it is equivalent to the effect of all other additive allelels at other loci. This results in continuous variation in F_2 but as we have seen in Fig. 12.9a, the transmission of genes still follows the Mendelian principles.

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12.10 PLEIOTROPY

We have seen cases, where one gene controls one phenotype. There are instances available, however, where one gene may control several phenotypes. Such a gene is known as **pleiotropic gene**. The basis of pleiotropy is the interrelationship between the metabolic pathways that may contribute towards different phenotypes. For example, mutations selected for one character may often affect several other characters. For instance, white eye mutation in *Drosophila*, lead to depigmentation in many other parts of the body, giving a pleiotropic effect.

SUMMARY

Genetics emerged as a formal discipline of biology only in the early twentieth century. Although its history is rich and dates back to pre-historic times, the observation that characters are passed on from generation to generation must have been made very early. The continuity of life and the variations therein, both form the integral components of the process of inheritance. The process of transmission of characters was established by Gregor Mendel when he studied the inheritance of seven characters in the garden pea plant. Mendel made certain postulates that led to the principles of inheritance. He showed that these characters are controlled by unit factors later called genes, that exist in pairs or alleles. Alleles exhibit dominance/recessiveness relationship with dominant allele able to express itself even in a heterozygous condition. Similarly, unit factors must segregate at the time of gamete formation so that each gamete receives one of the two unit factors with equal probability. Mendel also postulated that each pair of unit factors segregate independently of the other pair resulting in gametes with all possible combinations, with equal probability. The probabilities of phenotypes and genotypes are expressed in the form of genetic ratio. Mendel's work did not make any impact during his life time but the science of genetics was born in 1900 when these laws were independently discovered by three scientists.

After the rediscovery of Mendel's work, the study of genetics was expanded to include many other plants and animals and several other traits. This led to many alternative modes of inheritance. Basically, these arose due to two types of interactions, one at the level of alleles and the other between non-alleles. For example, incomplete or partial dominance and codominance led to different phenotypic expressions in the heterozygotes, owing to the absence of complete dominance. Unlike Mendelian concept of two alleles per gene, many genes were found to have multiple alleles. Such alleles generated by mutations can be detected only in a population, as a diploid organism can carry only two alternative alleles.

Modifications of Mendel's classical F_2 ratio were observed in several hereditary cases due to gene interactions. In epistasis, the expression of one gene masks the other gene or genes. The expressing or epistatic allele can be dominant or recessive over the one which gets masked and is called hypostatic. Some characters are also known to be controlled by several genes and such an inheritance is called the multiple gene inheritance. In such cases of polygenic or quantitative traits, several genes make additive contributions to phenotype and result into continuous variation. Single gene affecting the different characters which may be physiologically unrelated (pleiotropic genes) are also on record.

CHAPTER 13

CHROMOSOMAL BASIS OF INHERITANCE

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In Chapter 12, we have studied the transmission of characters from parents to offspring, which follows certain basic principles of genetics. In order to maintain the genetic continuity from cell to cell and from one generation to the next, in sexually reproducing organisms an orderly and coordinated process is required. This process must ensure the formation of haploid gametes, restoration of diploidy by fertilisation, and the generation and faithful multiplication of diploid somatic cells. The structure that serves as the vehicle of transmission of genes is chromosome. The manner in which the chromosomes and thus the genetic material, is transmitted from one generation of cells to another and from the organisms to their descendants is remarkably precise. This is accomplished by two cell divisional processes, mitosis and meiosis with which you have already been familiarised in Unit three. Although the two processes share many similarities the outcomes are very different. You may recall that mitosis faithfully duplicates and distributes the genetic material equally to the two daughter cells. On the other hand, in meiosis, the number of chromosomes is reduced to half.

In the previous Chapter, you have been familiarised with the principles of inheritance and the modifications in the phenotypic ratio that is brought about by various types of interactions both at the allelic and non-allelic levels. The importance of the principles of inheritance is also reflected by the fact that they are applicable to all living organisms including humans. We shall now look at inheritance on the basis of chromosomes. Though, inheritance will still be of the genes, it will be greatly influenced by their location on the chromosomes. The genetic studies done on the transmission of several allele pairs in different organisms also revealed that many times, genes do not follow the principle of independent assortment. In other words, they seem to be segregating *en block*. This tendency of genes to segregate together was referred to as linkage and it was later revealed that this characteristic could be assigned to genes located on the same chromosome.

Analysis of several genetic crosses done in different organisms also demonstrated that linkage between two genes, if located on the same chromosome, can be broken by the process of crossing over. Crossing over takes place during prophase I of meiosis, as you have studied in Chapter 11, and results in recombination or the formation of new combination traits. The incidence of recombination (in terms of per cent) is a good reflection of the distance between the genes in question and has been successfully employed to generate linkage or gene maps. In this Chapter, we shall familiarise ourselves with the concept of linkage and crossing over as well as the process of gene mapping.

While recombination is an important source of variation, the basic mechanism that brings about allelic variation is mutation. Alterations in chromosome number and structure also add to phenotypic changes.

13.1 PARALLELISM BETWEEN GENES AND CHROMOSOMES

In Chapter 12, you have learnt about how genes control phenotype and how genes are transmitted to the succeeding generations. A parallelism can be drawn between chromosomes and genes:

(i) Each species possesses a specific and characteristic number of chromosomes in its somatic cells. In a diploid cell, these chromosomes are found in pairs called homologous pairs which resemble in their

morphology and genetic content as we shall discuss later. One member of a homologous pair is derived from maternal parent and the other comes from the paternal parent. Similarly, each gene also exists as an allelomorphic pair which are contributed equally by the two parents.

(ii) Each chromosome is distinctly a double structure consisting of two sister chromatids. During mitosis, the two chromatids get separated into two daughter nuclei and then cells. Each member of an allele pair also finds a place in every daughter cell during mitosis. Such a distribution maintains the genetic composition of the cells constituting a multicellular organism.

(iii) At meiosis the members of a chromosome pair are separated from each other to go to daughter nuclei. Similarly, members of an allele pair also separate during meiosis so that each haploid cell or gamete contains only one allele.

(iv) When two haploid gametes unite during fertilisation not only the diploid chromosome number but also every allele pair is restored.

13.2 CHROMOSOME THEORY OF INHERITANCE

By the time Mendel was finalising his work on pea, chromosomes were seen in the nuclei of Salamander cells by William Flemming in 1879. The work of many cytologists including Flemming clearly suggested the role of nucleus in the process of inheritance. In 1902, cytologists, Walter Sutton and Theodor Boveri independently came to the conclusion that the behaviour of chromosomes at meiosis can serve as the cellular basis of both segregation and independent assortment. They, however, suggested that chromosomes can be equated with the Mendelian unit factors rather than genes located on the chromosome. Their work led to the chromosome theory of heredity, a concept which was further expanded by Thomas H. Morgan, Alfred H. Sturtevant and Calvin Bridges on fruit fly, *Drosophila*. The term **factor** as the basic unit of inheritance was replaced by Johannsen, in 1909, with the term **gene**.

Given the fact that the genes are located on the homologous pairs of chromosomes (Fig 13.1a), we can explain both the principles of segregation and independent assortment. When the members of a homologous pair separate, alleles carried on them will also segregate and go to different gametes. This can explain the segregation of alleles controlling the contrasting traits (Fig 13.1b). On the other hand, when we consider two homologous pairs each carrying an allele pair, one chromosome pair will segregate independently of the other, giving rise to four combinations that form the basis of independent assortment (Fig. 13.1c).

The phenotypic diversity that we see among the living organisms indicates that the number of genes is much more than the number of chromosomes. Therefore each chromosome carries a large number of genes. Whenever a gene is assigned a position on the chromosome, we call it a locus. Each allele pair though specifies the same character, it must carry slightly different information so as to reflect the contrasting forms.

13.3 INDEPENDENT ASSORTMENT VERSUS LINKAGE

You may recall the ***chromosome theory of heredity***. Here, the segregation and independent assortment of genes are explained on the basis of chromosomal segregation at Prophase I. We can assume two situations: either genes are located on different chromosomes or on the same chromosome. If you refer to Fig.13.1c, it will be clear that two genes located on different chromosomes always assort independently and thus show no linkage. In contrast, the genes located on same chromosome will present two situations: either a crossing over between the two genes or no crossing over occurs between the genes. Figure 13.2 illustrates the two situations and the consequence thereof. For example, when two non-sister chromatids undergo an exchange or crossing over, two new allele combinations can be generated. During meiosis I, when the crossing over takes place, each member of a homologous pair consists of two sister chromatids. You must also take note of the fact that only two non-sister chromatids participate in crossing over at any given point. This will lead to the formation of two types of gametes: those not involved in exchange will result into non-crossover or the parental type of gametes and those resulting from crossing over

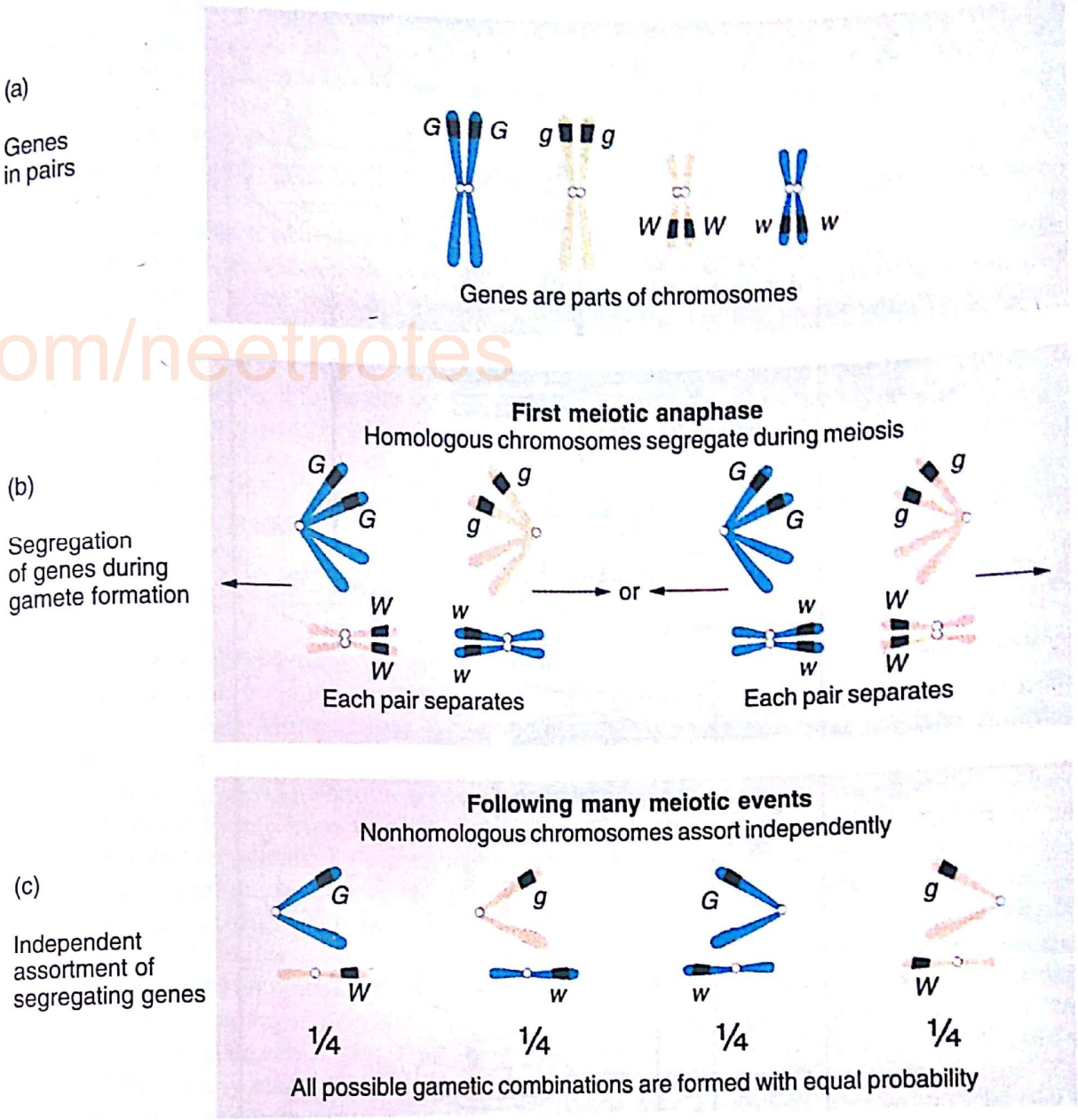

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Fig 13.1 *The correlation between the Mendel's laws of inheritance and chromosomal behaviour (a) Two pairs of genes G/g and W/w are depicted, located on two different homologous chromosome pairs (b) Segregation of each allele pair during gamete formation (c) Segregation of both the allele pairs assorting independently and producing all possible gametic combinations with equal probability*

are the recombinant types. If there is no crossing over between the genes, these will be transmitted intact. This will lead to the formation of only parental or non-crossover gametes and the phenomenon is known as **complete linkage** (Fig. 13.2). The frequency of crossing over between any two genes is generally proportional to the distance between them on the chromosome.

13.4 COMPLETE AND INCOMPLETE LINKAGE

As shown in Fig. 13.2, when we consider two genes located on the same chromosome, the situation can be explained if the two genes are very far from each other, so that crossing over is possible in all the meiotic cells. You may note that in such a case 50 per cent of parental and 50 per cent of recombinants

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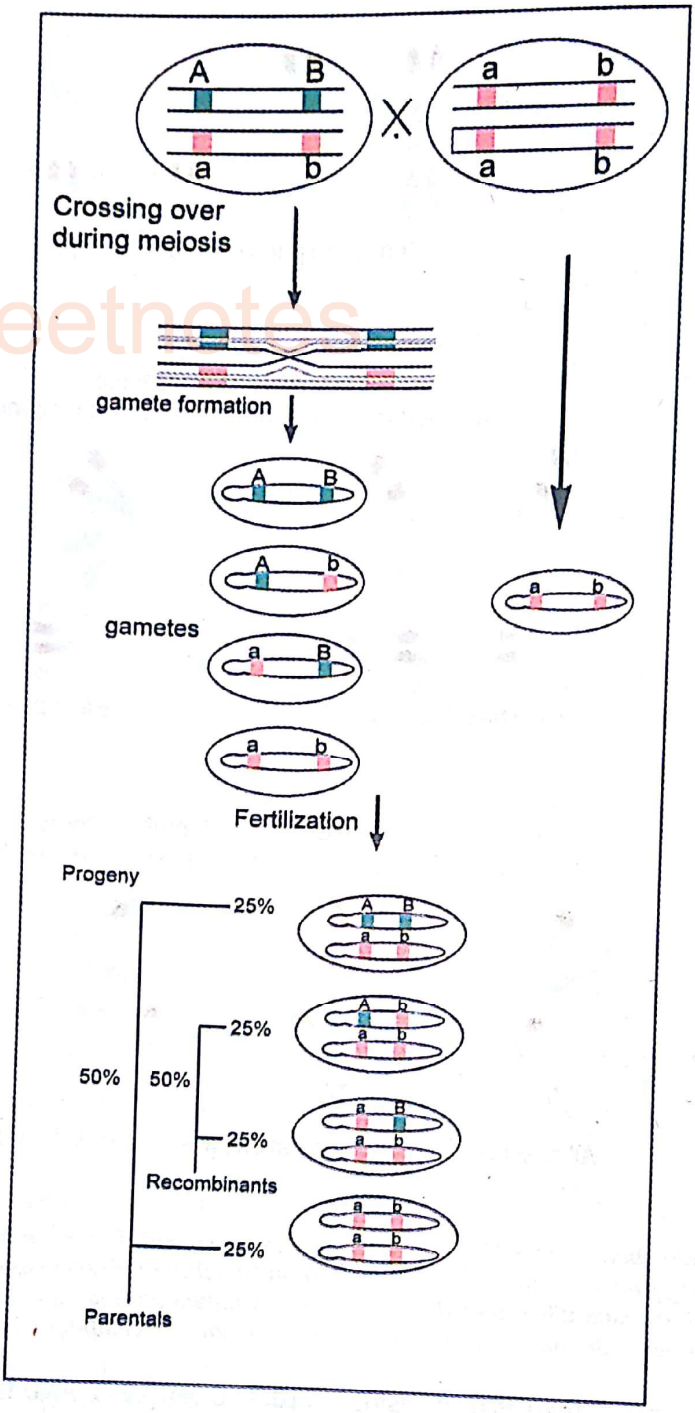

Fig 13.2 *Segregation of two allele pairs located on same chromosome, when they are far apart, will follow independent assortment*

will be produced and so the gametes will appear in a genotypic ratio of 1:1:1:1. In such an event, transmission of the two genes can not be distinguished from that of two genes located on different chromosomes and thus assorting independently. However, even when

the two genes are close to each other, crossing over does take place. In such cases incomplete linkage, there is a clear predominance of parentals or non-crossover types of gametes. This relationship was first demonstrated in *Drosophila* by Morgan and his student Sturtevant in 1911, and is still being upheld. From this discussion, it should be clear that all the genes located on the same chromosome will show some linkage between them. The strength of linkage will be determined by the distances between them; as farther they are, linkage can be easily broken by crossing over. Genes located on the same chromosome thus constitute a linkage group and the number of linkage groups in any organism is proportional to its haploid chromosome number.

13.5 CROSSING OVER

In 1910, Morgan started working on the fruit fly, *Drosophila melanogaster.* It could be grown on a simple synthetic medium in the laboratory. It can complete its life cycle in about two weeks, and a single mating could produce a large number of progeny flies. Thus, this fly had distinct advantages over pea plant. For example, pea plant will take a minimum of one season to produce seeds, which then have to be grown in the next season to check for some phenotypes such as plant height, flower colour, etc. Further, there is a clear differentiation of male and female flies unlike the bisexual flowers in pea. Thus, no selfing is possible at any stage; instead female and male flies of desired genotypes can be interbred.

Morgan made several monohybrid and dihybrid crosses in *Drosophila* and deduced important genetic information. For example, in one of his dihybrid crosses involving yellow body (y) and white-eyed (w) mutant females and wild-type males (grey body and red eye), the F_1 females were wild-type and males expressed both the mutant characters (Fig.13.3). When such F_1 females and males were crossed, the F_2 progeny consisted of 98.7 per cent yellow-bodied and white-eyed flies, as well as wild-type flies. The remaining 1.3 per cent consisted of yellow-bodied and red-eyed flies, as well as grey bodied and white-eye types. If we classify them on the basis of their phenotypes, the former class is the parental type and the latter is the recombinant type, where new combinations of genes have been produced through an exchange that has occurred in F_1 flies during gamete formation. In a similar cross involving white eye and miniature wings, Morgan found 62.8 per cent F_2 offspring having the parental phenotype and 37.2 per cent, where new gene combinations had occurred.

In 1911, Morgan was confronted by two main questions:

(i) how do the genes get separated? and

(ii) why did the frequency of apparent separation differed between different genes?

Morgan's knowledge of F. Janssens's cytological observations came handy in providing an explanation for the same. Janssens had shown that synapsed homologous chromosomes during meiosis appear to be engaged in an exchange through **chiasmata** (*sing.* chiasma). Morgan proposed that these chiasmata could represent the points of crossing over, or genetic exchange that leads to recombination. From the results of various crosses, Morgan concluded that linked genes are arranged linearly on the chromosome and two genes can get separated if an exchange occurs between them.

13.6 RECOMBINATION

We have already introduced the term recombination and have widely used this term to refer to new gene combinations. It is, however, absolutely necessary to understand this term clearly. **Recombination** is a process that can occur in a variety of situations but the most popular context is in relation to meiosis. Thus, this is a meiotic process that generates haploid product whose genotype is different from the two haploid genotypes constituting the

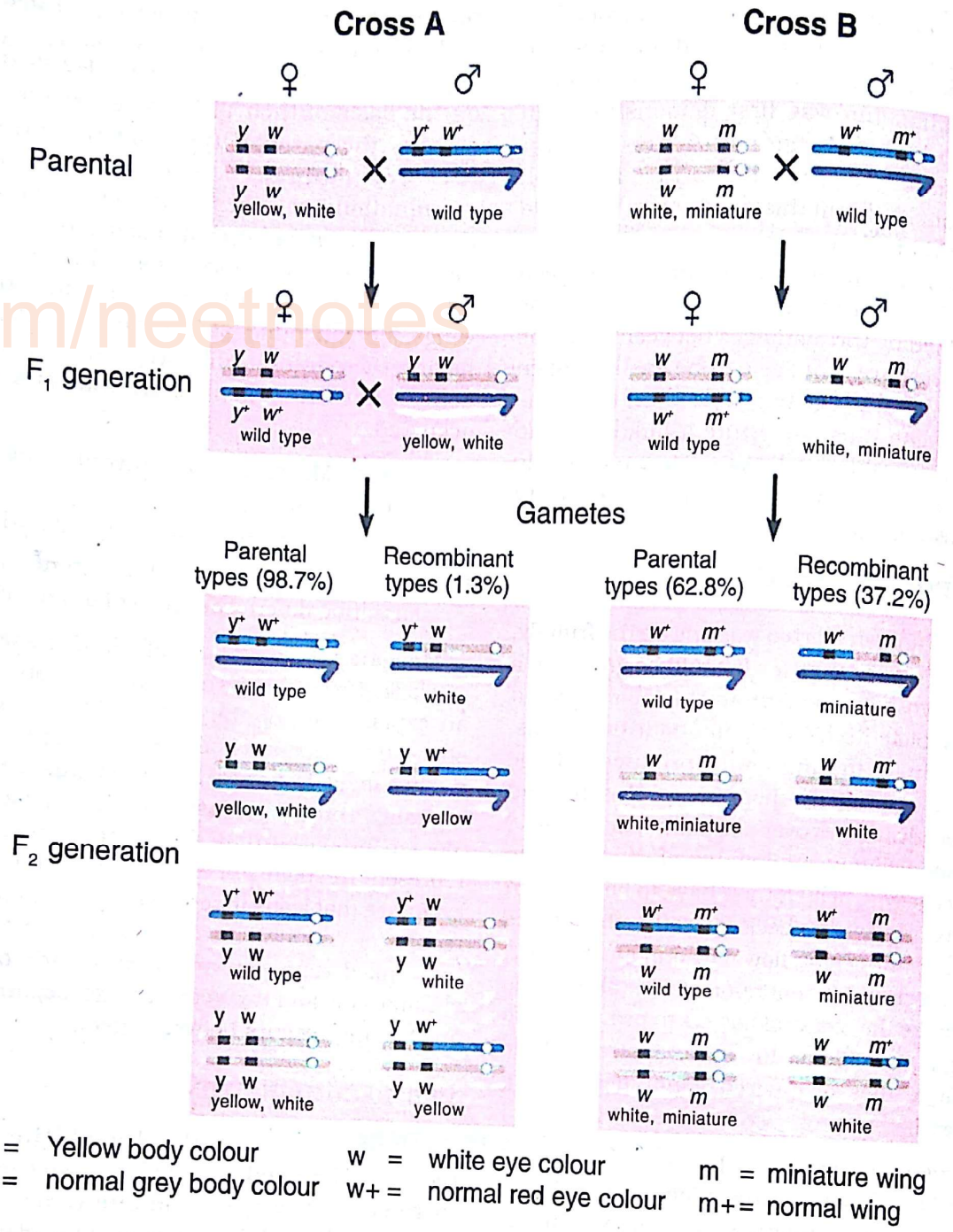

Fig 13.3 *Results of two dihybrid crosses conducted by Morgan. Cross A showing crossing between gene y and w; Cross B showing crossing between genes, w and m.*
Note: The strength of linkage between y and w is higher than w and m

diploid. The product of recombination is a **recombinant** (Fig.13.4).

You may recall that Morgan had suggested that the formation of recombinants can be explained by the occurrence of crossing over (chiasma formation) during meiosis I. This led to what is

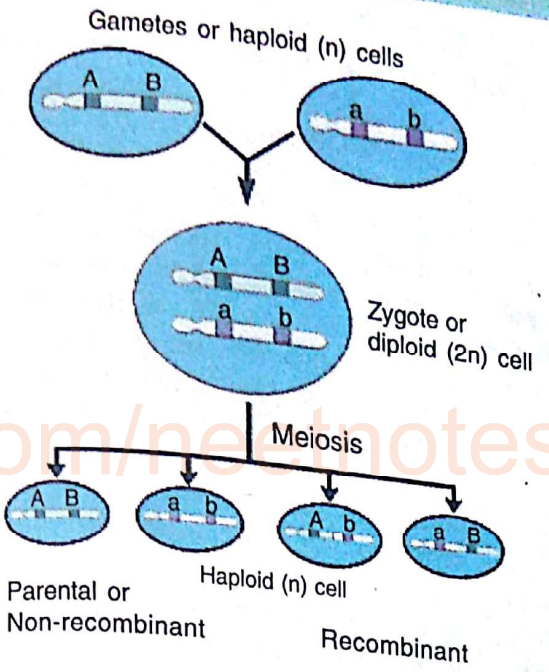

Fig 13.4 *Formation of recombinant as well as non-recombinant (parental type) gametes*

known as **chiasma-type hypothesis**, which is based on the following (Fig.13.5):

(i) During prophase I (zygotene), homologous chromosomes pair (synapsis), with each chromosome consisting of two chromatids.

(ii) During pachytene, two non-sister chromatids break at homologous sites and then rejoin.

(iii) This rejoining takes place between non-sister chromatids leading to physical exchange.

(iv) The physical exchange leads to reshuffling of genes and thus recombination.

13.7 CHROMOSOME MAPPING: A STEP TOWARDS A GENE MAP

The gene mapping exercise requires the setting up of a test cross and the estimation of recombination frequency between the required genes. The latter is determined by the relationship:

$$\text{Recombination frequency} = \frac{\text{Total number of recombinarts}}{\text{Total number of progeny}}$$

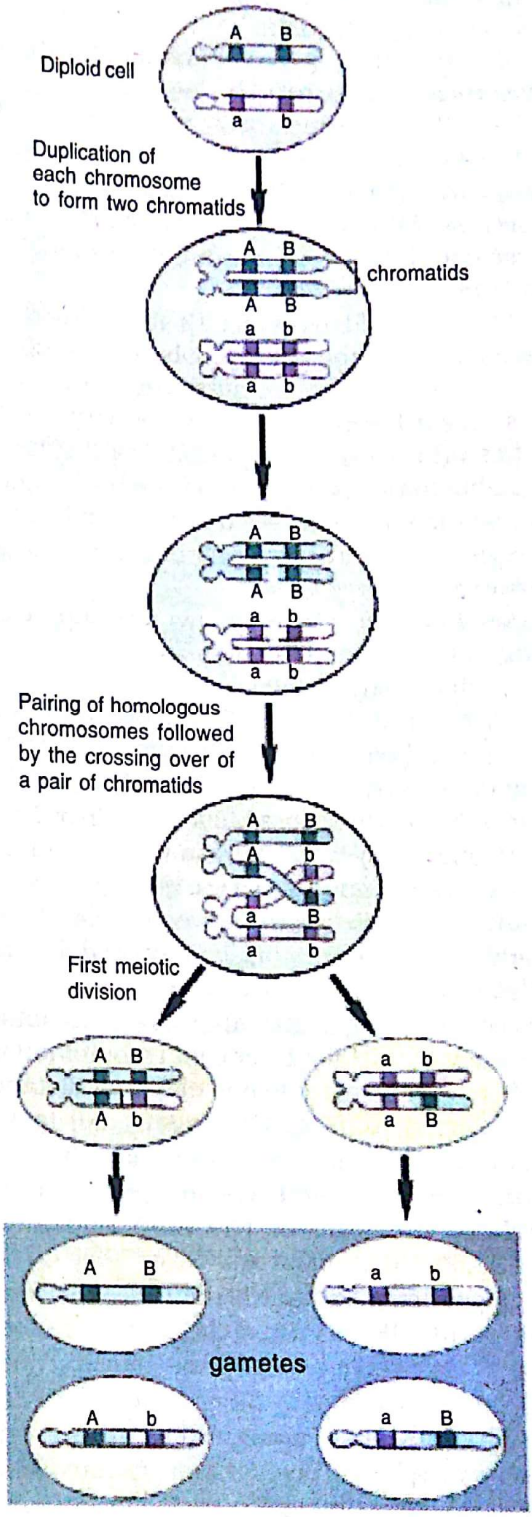

Fig 13.5 *Formation of chiasma and resulting in recombinant formation. Breakage and reunion of non-sister chromatids lead to shuffling of genetic contents or recombinants*

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The value is generally represented as a per cent of the total population.

The variation in recombination frequency is governed by the distance between the genes. Closer the two genes, less are the chances of crossing over. This is reflected in the lower frequency of recombination and vice-versa. In contrast, if the two genes are far apart, crossing over could occur at many points (double or multiple crossovers).

Alfred H. Sturtevant first realised the importance of these proposals and suggested that the frequency of recombination can be used to suggest the relative distance between the genes on the chromosome. It could thus, be possible to produce a map of the linked genes (linkage map). Let us see how this can be done. Frequencies of crossing over or recombination between the three genes:
yellow body (y), white eye (w) and miniature wing (m), studied by Morgan are:

Yellow body – white eye 1.5%,
White eye – miniature wing 34.5%,
Yellow body – miniature wing 36.1%.

How do we arrive at a sequence of these three genes? In fact three possibilities exist in ordering these genes: (i) y w m , (ii) y m w, (iii) w y m.

Sturtevant argued that if the genes are linearly arranged, the distances between them should be additive. Based on this, he proposed the map of these genes (Fig. 13.6 a, b).

In constructing such a map, a recombination frequency of 0.01 (or 1 per cent recombination) is taken as equivalent to one map unit distance now referred to as **centimorgan** (cM) in the honour of Morgan's contributions. The map distance between y and m is 36.1 per cent units which is the sum of the map distances 1.5 + 34.5 = 36 (in most real cases, they do not add up perfectly, because of double crossovers).

Different alleles of a gene act as genetic markers that serve as signposts to help reveal the relative locations of other genes along the chromosome. The assignment of genetic markers to relative positions on chromosome produces a **genetic map** of that chromosome (Fig.13.6c).

In recent years, the most detailed genetic maps have been generated by determining the nucleotide sequence of the DNA that makes up each chromosome.

Why did Mendel not detect Linkage?

As we have discussed, two genes located on the same chromosome may not assort independently and exhibit linkage. We have seen in Chapter 12 that Mendel worked with 7 pairs of alleles in pea and did not report any linkage. In other words, none of the allele combinations gave results which could not be explained on the basis of independent assortment. It was initially thought that Mendel selected seven characters and there are seven pairs of chromosome in each diploid cell in pea. Thus these genes that could have been distributed on different chromosomes, exhibited independent assortment, and their selection was a matter of luck. Detailed investigations by S. Blixt on pea led to the location of Mendel's seven selected characters on four different chromosomes: two on chromosome 1, three on chromosome 4 and one each on chromosomes 5 and 7. Of the two genes on chromosome 1, they are so distantly located that no linkage is normally observed. Similarly, two out of the three on chromosome 4 are very far in relation to the third to show any linkage. This leaves only one gene pair: full vs constricted pod and tall vs dwarf plant height which ought to have shown linkage.

If one goes through Mendel's published data, this particular combination has not been mentioned at all, presumably because such a cross was never made. It thus explains as to why Mendel did not run into the problems of linkage.

Whenever a new variant gene is discovered, one of the first questions to be asked is 'where is it located?' This mapping is not only important in genetic engineering, but is also of fundamental importance in understanding the overall architecture of the chromosome and in fact of the entire genome.

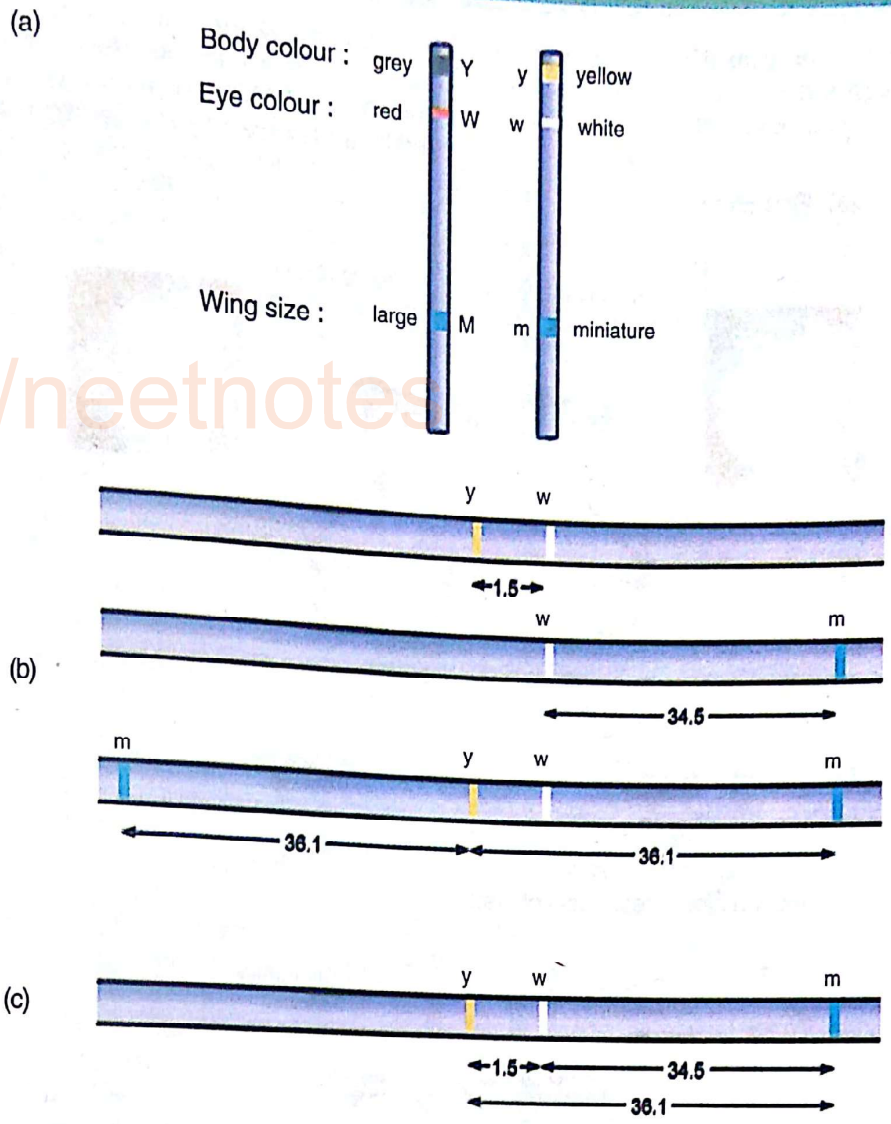

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Fig 13.6 *A chromosomal map (linkage map) in Drosophila (a) The region showing the relative location of the genes y, w and m (b) represents the map distance between y and w ; and w and m; and the possibility of placing m to the left or right of y-w (c) The actual map derived from the same*

13.8 SEX-LINKED INHERITANCE

In many sexually differentiated (dioecious) organisms, each cell contains a pair of chromosomes known as **sex chromosomes**. In many animals and plants, one of the sexes contains a pair of unlike chromosomes and the other sex contains a pair of similar chromosomes, the sex chromosomes. For example, both in humans and *Drosophila* the males contain X and Y and the female XX chromosomes. To distinguish this pair of sex chromosomes, all other chromosomes are referred to as **autosomal chromosomes** or simply **autosomes**. We have also seen earlier that genes located on the same chromosome show linkage. We shall now follow the inheritance of a trait which is linked to sex chromosomes and see whether a difference exists. One of the first mutant allele studied by Morgan in 1910 for his work on linkage in *Drosophila*, in fact, was located on the X-chromosome. This mutant allele giving white eye (w) phenotype was recessive to its wild-type red eye (w^+) allele. Morgan's work established that the inheritance of white-eye trait is related to the sex of the parent carrying the mutant

allele. We shall learn about this concept by looking at the results of a reciprocal cross involving white-eyed and red-eyed *Drosophila* (Fig.13.7).

If you look at the results of these monohybrid crosses, they are not identical. You may recall that in all of Mendel's monohybrid crosses, F_1 and F_2 data were very similar regardless of which

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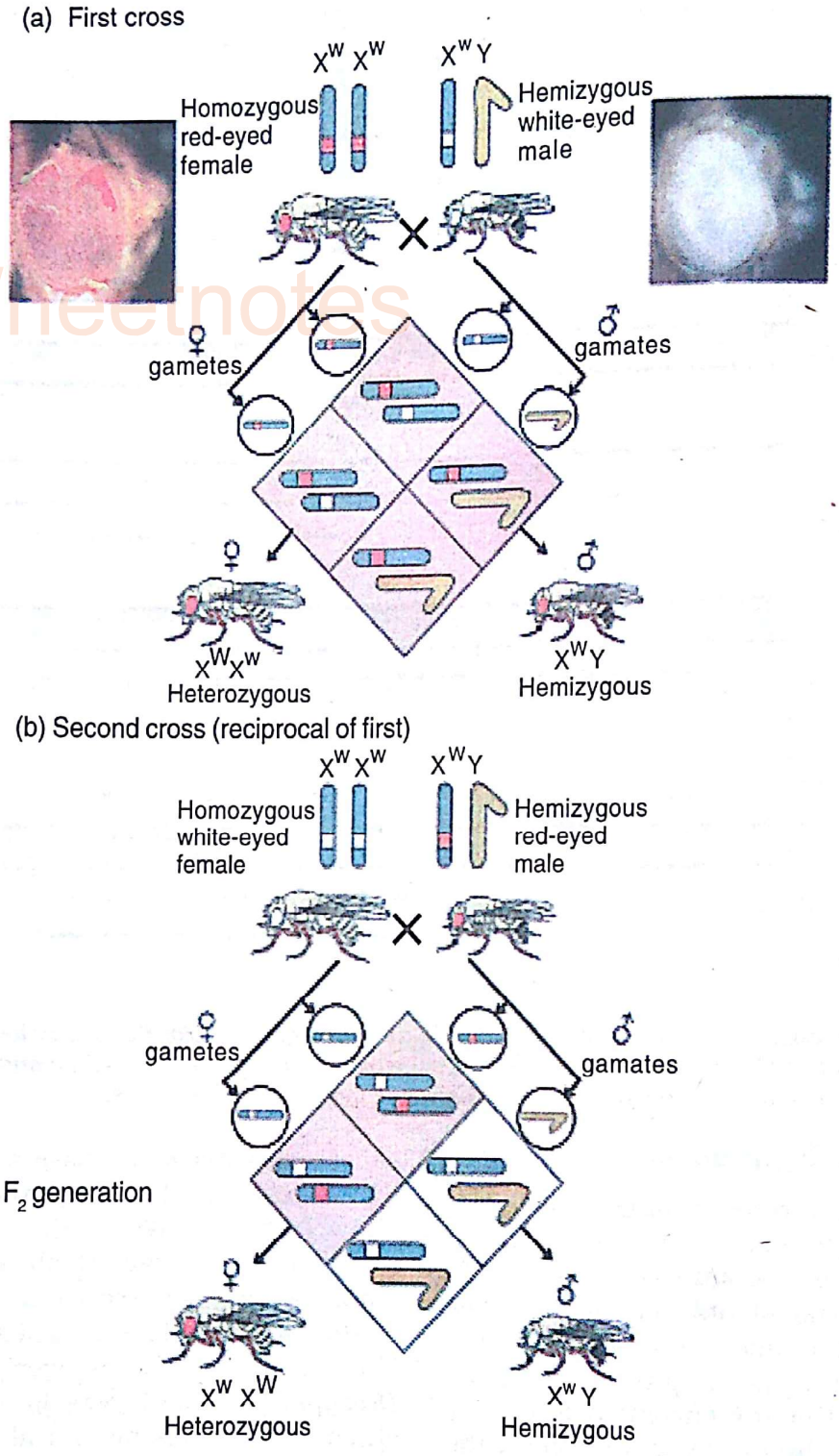

Fig 13.7 *Inheritance of a sex-linked character. Inheritance of white-eye allele located on X chromosome in Drosophila (a) Female carry red-eye and male white-eye allele (b) A reciprocal cross where white-eye allele is contributed by female and male carries the red-eye allele. Note that the outcome of a reciprocal cross is different*

parent contributes the recessive trait. In contrast, Morgan suggested that X-linked recessive allele shows a criss-cross pattern of inheritance, i.e. recessive genes are transmitted from homozygous mothers to all the male offspring. He hypothesised that the males carry only one allele and that too on X chromosome but no corresponding allele on Y, (a situation referred as hemizygous) while females with 2X chromosomes carry two alleles like any other homologous pair of chromosomes.

Morgan's work on sex chromosome, and that of his student Calvin Bridges, provided further support to chromosome theory of inheritance.

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13.9 BASIS OF SEX DETERMINATION

We have seen above that both *Drosophila* and human chromosome complements consist of autosomes (2A) and sex chromosomes; female with 2A+XX and male with 2A+XY. In humans and *Drosophila*, the male produces two types of gametes while the female produces only one type of gamete (Fig. 13.8a, b). Therefore in human beings, the sex determination of the progeny is determined by the male gamete at the time of fertilization. Subsequent studies have established that Y chromosome carries a gene *Sry* (Sex determining region) which codes for a product called testis-determining factor (TDF). TDF is required for the development of male sex and its absence leads to the development of female sex. This observation might lead us to conclude that sex is determined by a pair of chromosomes or sex chromosomes and Y is responsible for maleness in both the species. Evidence gathered from study of variations in sex chromosome composition in these cases and subsequent molecular genetic analysis proved that Y does determine maleness in humans and other mammals, but not in *Drosophila*.

The work of Calvin Bridges on *Drosophila* demonstrated that in this fly, sex determining factor is the ratio of number of X chromosomes to the sets of autosomes. Thus, 2A+2X or 3A+3X having an X/A ratio of 1.0 are female and fertile. An X/A ratio of 0.5 is required for male sex differentiation. Bridges also showed that flies with X/A ratio between 0.5 and 1.0 exhibited a lot of morphological and sexual abnormalities. These sterile flies were referred to as intersexes. These results suggested that in *Drosophila* male

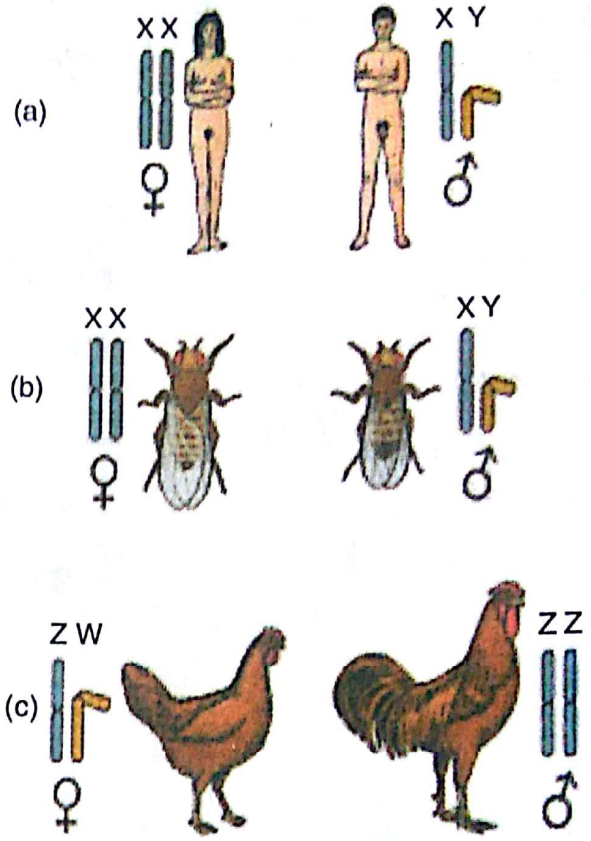
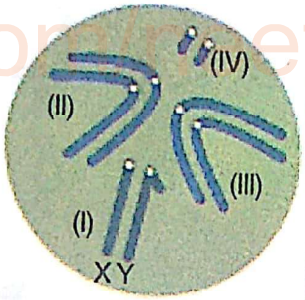


Fig 13.8 *Determination of sex by chromosomal differences (a,b) Both in humans and in Drosophila, the female has a pair of XX chromosomes (homogametic) and the male XY (heterogametic) composition (c) In many birds, female has a pair of dissimilar chromosomes ZW and male two similar ZZ chromosomes*

development factors are localised on autosomes instead of sex chromosomes. The X chromosome, however, does seem to carry some female determining genes such as *Sxl*. Such a mode of sex differentiation is referred to as the genic balance theory of sex determination (Fig. 13.9).

In birds and butterflies, males are XX and females are XY. To avoid confusion, these forms are usually expressed as ZZ (male) and ZW (female) (Fig 13.8c). In these organisms, the female produces two types of gametes. Thus, the egg determines the sex of the offspring.

Normal diploid male



Chromosome composition	Chromosome formulation	Ratio of X Chromosomes to autosome sets	Sexual morphology
	$3X/2A$	1.5	Metafemale
	$3X/3A$	1.0	Female
	$2X/2A$	1.0	Female
	$2X/3A$	0.67	Intersex
	$3X/4A$	0.75	Intersex
	$X/2A$	0.50	Male
	$XY/2A$	0.50	Male
	$XY/3A$	0.33	Metamale

Fig 13.9 *Genic balance concept of Sex determination in Drosophila as proposed by Bridges*

13.10 GENETIC VARIATION

We shall now return to a concept that we introduced in Chapter 12. You may recall that along with heredity there is always a component of variation. In other words, while a whole set of characters is transmitted from the parents to the offspring there is always a component of variation due to which they not only look different from one another but also the offspring can be distinguished from each other. These variations may arise due to different mechanisms, such as recombination, gene-mutation, gene-environment interactions and less frequently by chromosomal aberrations. We have already discussed the process of recombination; in the following subsections we shall be discussing the other two mechanisms.

Gene Mutation

The variations at the level of gene which form the basis of the differences between related organisms as well as the diversity among organisms is assigned to a process called **gene mutation**. The term mutation was first coined in 1901 by Hugo de Vries to explain the variations he observed in the plant evening primrose, *Oenothera lamarckiana.* However, most of these variations turned out to be chromosomal aberrations that we shall discuss later.

Mutation is of basic importance in genetics and is defined as *a sudden, discrete change in the genetic material (gene) which is heritable.* Mutation may often lead to phenotypic variations.

It is now clear that mutation serves as the source of most of the existing genetic variability in a population. This variability provides the raw material for natural selection and therefore, biological evolution. The resulting phenotypic variability also allows the geneticists to study the genes that control the modified trait and follow its transmission to the subsequent generations. Thus, mutations provide the 'markers' without which genetic analysis will be impossible. For example, if all the pea plants displayed the same phenotype, Mendel could not have conducted his experiments. Because of such contributions, great attention has been paid to the origin, classification, induction and application of mutations.

Mutations when they arise suddenly in nature, are called **spontaneous mutations**. However, many agents such as radiations and some chemicals, called **mutagens** can be applied to create **induced mutations**. Occurrence of mutations in gametic cells can get transmitted to next generation but those in somatic cells are confined only to that individual. Similarly, in a diploid cell a recessive mutation will not be expressed, as it may be masked by its dominant wild-type allele. In order to express, it ought to be either in a homozygous state or, a dominant autosomal or a X-linked recessive mutation. Similarly, mutations occurring in early developmental stages have better chances of spreading than those in adult tissues. While mutations affecting a morphological trait are the easiest to detect, nutritional or biochemical, behavioral, regulatory and lethal mutations can also be induced. We shall discuss the molecular mechanism of mutation in Chapter 14.

Chromosomal Aberrations

Besides gene mutations, alterations in the chromosomes can also occur. Since these changes may bring about visible changes in the phenotype, they have been referred to as **chromosomal mutations** or **chromosome aberrations**. You may recall that in gene mutations, no change in the chromosome occurs. These chromosome aberrations can either involve a change in the structure of the chromosome, or a change in the chromosome number.

Changes in Chromosome Structure

You have studied in Unit Three and the early part of this Chapter that every living cell possesses a specific set of chromosomes which bear a specific size and structure, and occur in pairs in a diploid organism. Any alteration in the chromosome architecture can be detected either by genetic tests or cytologically by simply viewing the chromosomes, preferably in paired condition under the microscope. Four different types of structural alterations (deletion, duplication, inversion, translocation) have been identified all of which involve chromosome breakage and then rearrangements in some cases. Many of these aberrations change the order of genes on the affected chromosome.

Sometimes a part of the chromosome is lost. This is known as **deletion** or deficiency. This loss can be from one end or from chromosome parts between the ends. Accordingly, the former is known as **terminal** and the latter **interstitial deletion** (Fig 13.10a). For example, the Cri-du-chat syndrome is caused by the loss of half of the short arm of chromosome 5 in humans. In this disorder, infant's cry resembles the sound of a cat experiencing pain.

A **duplication** occurs when a portion of a chromosome is repeated. Aquisition of additional genetic material can lead to new functions or phenotype or serious deleterious effects, e.g. the development of bar eye in *Drosophila* (Fig 13.10b).

When a segment of chromosome breaks but later rejoins after rotating by 180° it results in **inversion**. If the centromere is included in the inverted segment, it is known as **pericentric** but if inversion occurs only in one arm and the centromere is not involved, it is referred as **paracentric** inversion (Fig 13.10c). As many as 1 per cent newborns may carry an inversion that can be detected by G-banded chromosome karyotyping.

Sometimes a segment of chromosome breaks and relocates within a non-homologous chromosome. This leads to **non-reciprocal translocation** (Fig. 13.10d). In another type, two non-homologous chromosomes exchange a segment; this creates a **reciprocal translocation**. The latter was the event referred by Hugo de Vries in 1901 in evening primrose.

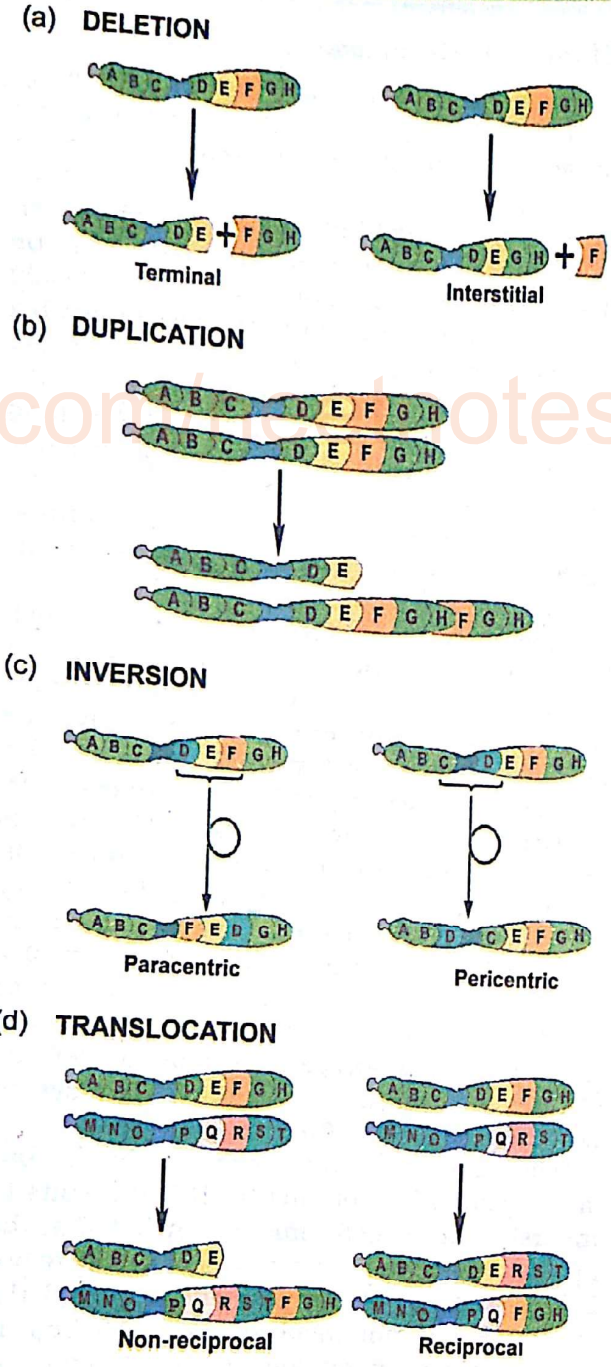


Fig 13.10 *Four different types of aberrations arising by changes in the chromosome structure/ architecture*

We shall also refer to translocations later in relation to Down's syndrome. In certain leukemia, such as chronic myloid leukemia (CML), the malignant cells have the chromosome 22 shortened due to translocation of a piece of its long arm. Inversions and deletions produce unbalanced meiotic products thus leading to sterility. Some of them cause new location for a gene and so may often lead to changed expression.

Changes in Chromosomal Number

Like structure, a change in chromosome number can also bring about visible effects on the phenotype. You already know that each species has a characteristic number of chromosomes which is a reflection of a basic set or **monoploid** number (x). Those having multiples of monoploid number are **euploids** and those euploids which have more than twice the number of monoploid are called **polyploids**. Thus, besides monoploid (x) and diploid (2x) polyploid series consists of triploid (3x), tetraploid (4x), pentaploid (5x), hexaploid (6x), etc. Organisms with changes that involve individual chromosomes are referred to as **aneuploids**.

Among polyploids, the increase in chromosome number may involve the same set of chromosomes within a species (**autopolyploid**) or may consist of sets from different species (**allopolyploid**). The two classes are referred by prefix auto- and allo-, for example, autotetraploid and allotetraploid, etc. Many of our important crop plants such as wheat, cotton and tobacco are natural allopolyploids, and allopolyploidy has resulted into first man-made cereal, **Triticale**.

Aneuploidy may be a case of loss of one (2n-1) or both the homologous chromosomes (2n-2). The former is called **monosomic** and the latter **nullisomic**. Similarly, gain of a chromosome (2n+1) is referred to as **trisomic** and so on. We have referred to such situations while discussing human disorders later in this Chapter.

Polyploidy has important applications in crop breeding and horticulture. More so, allopolyploidy can be used to pool genetic information from two distinct species to create a new species. Aneuploidy has also been used in crop engineering. In humans, aneuploidy has led to the unravelling of roles of certain chromosomes (both autosomes and sex chromosomes) in normal human development.

13.11 PROKARYOTIC CHROMOSOME

Prokaryotic cell lacks nuclear membrane and thus the genetic material is found in a compact structure called **nucleoid**. The nucleoid consists of generally a large circular chromosome approximately 1.2mm in length in the bacterium, *Escherichia coli*. The chromosome is made up of DNA with associated proteins. These proteins have close similarities with histones found in eukaryotes. Also a large proportion of positively charged ions of proteins help them to counter balance the negative charge of phosphate groups in DNA.

The large size of the chromosome in comparison to that of the cell demands coiling and supercoiling which can be easily visualised by electron microscope when the DNA is released by gentle lysis of the cell. Thus, a chromosome is a covalently closed circular structure,

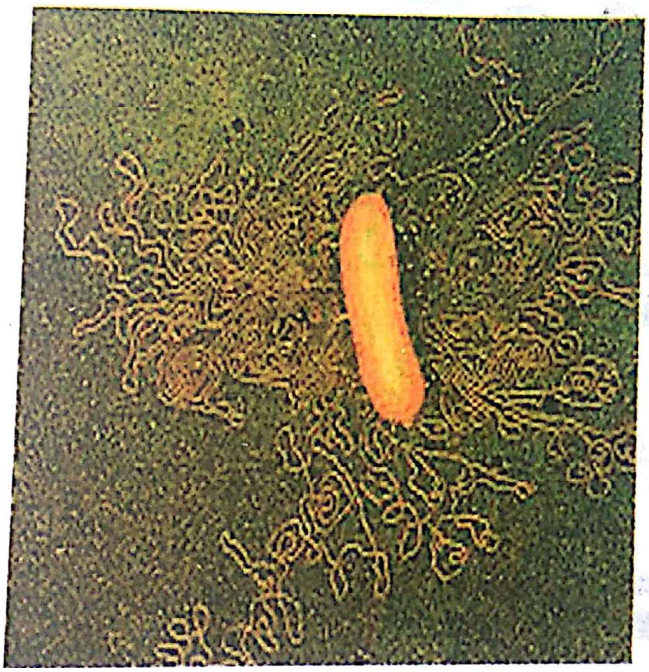


Fig 13.11 *A photomicrograph of a bacterial cell lysed to release the nucleoid. Note that it is a highly coiled structure*

consisting of several loops of supercoiled DNA (Fig. 13.11).

13.12 EUKARYOTIC CHROMOSOME

Unlike prokaryotes, eukaryotic chromosome structure and its organisation is much more intricate. This complexity is because of greater amounts of DNA per chromosome, more number of chromosomes and large number of proteins associated with DNA. In comparison to *E. coli* chromosome of about 1.2 millimetre, the 46 human chromosomes taken together will measure up to 2 metres. Such a large amount of chromatin material is located inside a nucleus which is no bigger than 5µm in diameter. So the chromatin condenses almost 10,000 times to give the size of a mitotic chromosome. In eukaryotic chromatin, DNA is associated with positively charged histones and less positively charged non-histone proteins.

The association of histones with the DNA is very characteristic. It involves the formation of linear arrays of spherical structures known as **nucleosomes**. Each nucleosome consists of an octamer of histones. A length of DNA is wrapped around these histones to produce the nucleosome core particles. Histone H_1 is situated outside of the nucleosomal DNA in the linker region. The nucleosomal organisation provides a chromatin fibre approximately 10nm in thickness, which gets further condensed to produce a solenoid of a 30 nm diameter. This solenoid structure undergoes further coiling to produce a chromatin fiber of 300 nm and then a chromatid of 700 nm diameter which can be seen under the light microscope (Fig.13.12). All the folded loops of chromatin are held by a nuclear scaffold formed by non-histone proteins.

13.13 HUMAN GENETICS

We, like all other living organisms, also follow the principles of inheritance. One of the biggest difficulty encountered is that controlled crosses cannot be made in human inheritance studies. Also the number of offspring produced per mating is very restricted unlike many other organisms such as pea plant.

Methods of Study

1. Karyotyping: see chapter 9, Fig .9.18
2. Pedigree Analysis: A family tree or pedigree is drawn for families having genetically transmitted diseases or traits. This is constructed based on information gathered about all members of the family over many generations. This pedigree is represented with

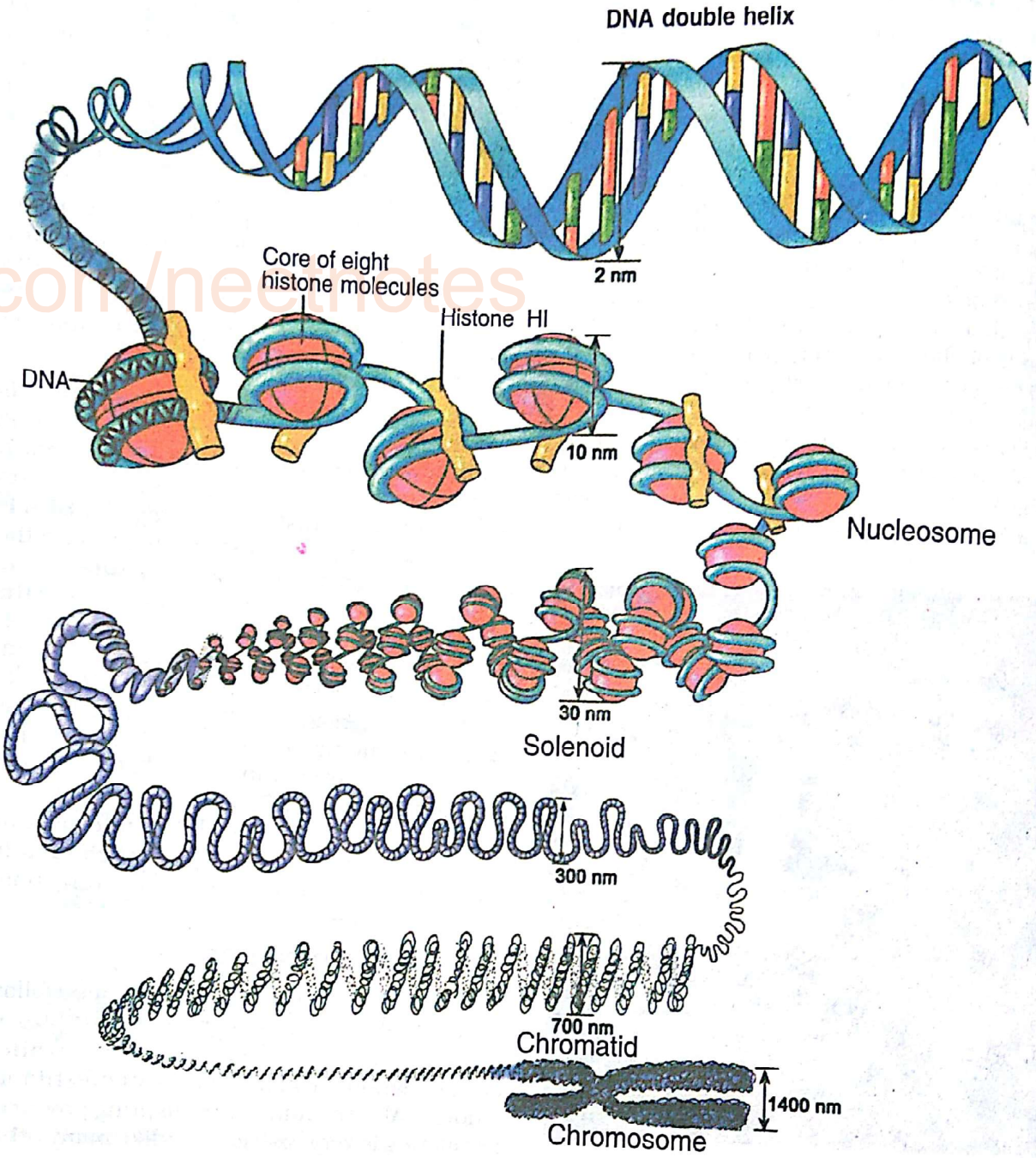


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Fig 13.12 *Various steps in the folding and super folding of the basic chromatin components to generate an eukaryotic chromosome*

certain standard symbols. Some of these symbols are given in Fig. 13.13.

For carrying out simple Mendelian analysis involving recessive or dominant allele, certain clues or simple rules are sought from the pedigree. In the case of recessive allele, for example, characteristic condition can appear in the progeny of apparently unaffected parents.

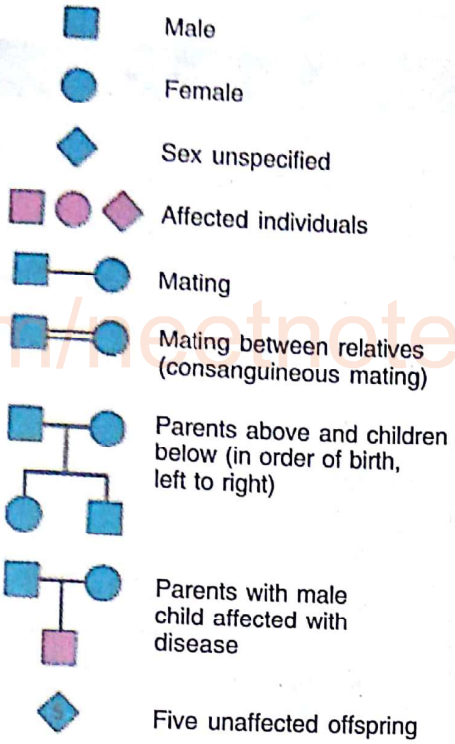

Fig 13.13 *Symbols used in human pedigree analysis to study the inheritance of a character*

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Moreover, two affected individuals cannot have unaffected child. Quite often, such recessive alleles are revealed by close marriages, such as cousin marriages.

Simple pedigree analyses have extensive use not only in medical research but also in the day-to-day counselling of prospective parents who would like to be guided about the possibilities of transmitting a diseased condition to their children. Very often a single pedigree analysis does not offer the conclusions that can be drawn from a designed cross producing large number of offspring. In such case, several independent pedigrees involving the same trait are analysed so as to draw consistent conclusions (Fig.13.14).

Genetic Disorders

Interest in human heredity arose since very early times and logically so, as human beings follow the same phenomenon of transmitting the characters to their progeny and generating variations. However, serious genetic analysis began soon after the rediscovery of Mendel's laws of inheritance. In 1902, Sir Archibald Garrod and William Bateson reported several disorders

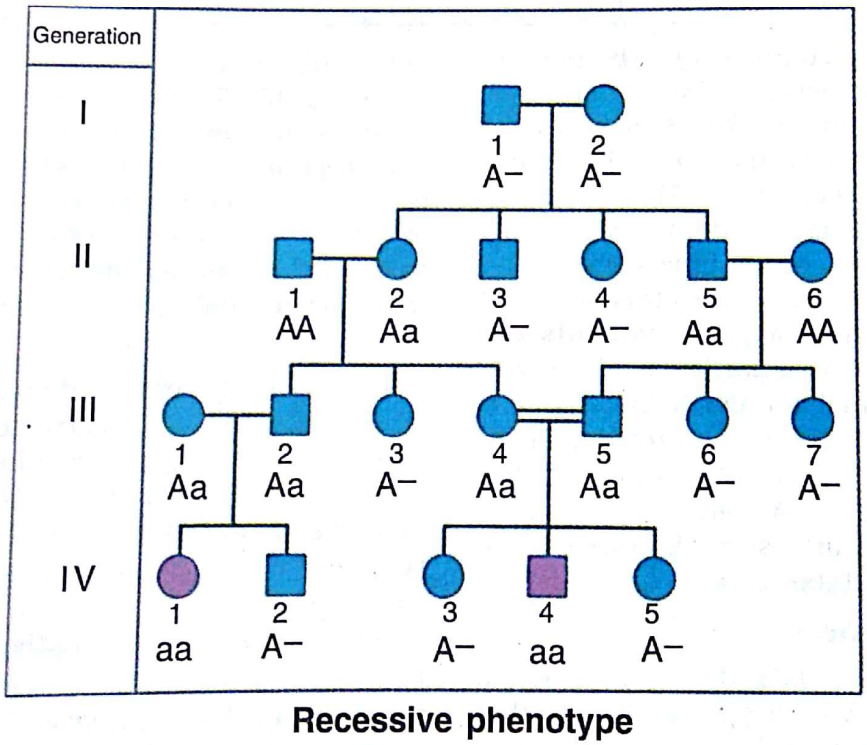

Fig 13.14 *Illustrative pedigree, involving the inheritance of a recessive allele determined phenotype*

Table 13.1. Some Human Genetic Disorders

Disorder	Dominant/ Recessive	Autosomal/ Sex linked	Symptom	Effect
Sickle-cell anaemia	Recessive	Autosomal (Chromosome 11)	Aggregation of erythrocytes, more rapid destruction of erythrocytes leading to anaemia.	Abnormal haemoglobin in RBC's
Phenylketonuria	Recessive	Autosomal (Chromosome 12)	Failure of brain to develop in infancy, mental retardation.	Defective form of enzyme phenylalanine hydroxylase
Cystic fibrosis	Recessive	Autosomal (Chromosome 7)	Mucus clogging in lungs, liver and pancreas anomalies.	Failure of chloride ion transport mechanism.
Huntington's disease	Dominant	Autosomal (Chromosome 4)	Gradual degeneration of brain tissue in middle age	Production of an inhibitor of brain cell metabolism
Haemophilia A/B	Recessive	Sex-linked (X chromosome)	Failure of blood to clot	Defective form of blood clotting factor VIII/IX.
Colour blindness	Recessive	Sex-linked (X chromosome)	Failure to discriminate between red and green colour.	Defect in either red or/and green cone

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in human population that seemed to be inherited like any Mendelian gene. In many such cases, the disorder could be traced to a step in metabolism (we shall discuss one such disorder, alkaptonuria, in Chapter 14). These errors or disorders are transmitted to the progeny following the same laws of inheritance as we have discussed earlier. These, therefore, not only revealed, how a gene controls the metabolism and thus a particular phenotype, but its pedigree analysis also helps in genetic counselling. Genetic counselling is an important tool to check the spread of such a diseased condition. We shall use certain examples to look into some known human genetic disorders (Table 13.1).

Sickle-cell Anaemia

Sickle-cell anaemia is a disorder in which afflicted individuals contain red blood cells that under low oxygen tension become elongated and curved (sickled). Such sickling of erythrocytes does not occur in normal individuals and they retain a biconcave disc shape (Fig. 13.15a). Individuals with this disease suffer attacks due to aggregation of red blood cells in the capillary systems of venous side and several tissues suffer severe damage due to paucity of oxygen, also known as sickle-cell crisis. Also, these erythrocytes are destroyed more rapidly than the normal red blood cells leading to anaemia.

The disease is controlled by a single pair of alleles, Hb^A and Hb^S. Pedigree analysis revealed three genotypes and two phenotypes with normal and affected individuals resulting from homozygous genotypes Hb^AHb^A and Hb^SHb^S respectively and hetrozygote Hb^A Hb^S exhibiting sickle-cell trait. Though heterozygotes are apparently unaffected, as they carry atleast one normal allele, they are carriers of the defective gene and can transmit the same to 50 per cent of their offspring, on an average (Fig.13.15b). This defect arises from the substitution of valine (Val) for glutamic (Glu) acid at the

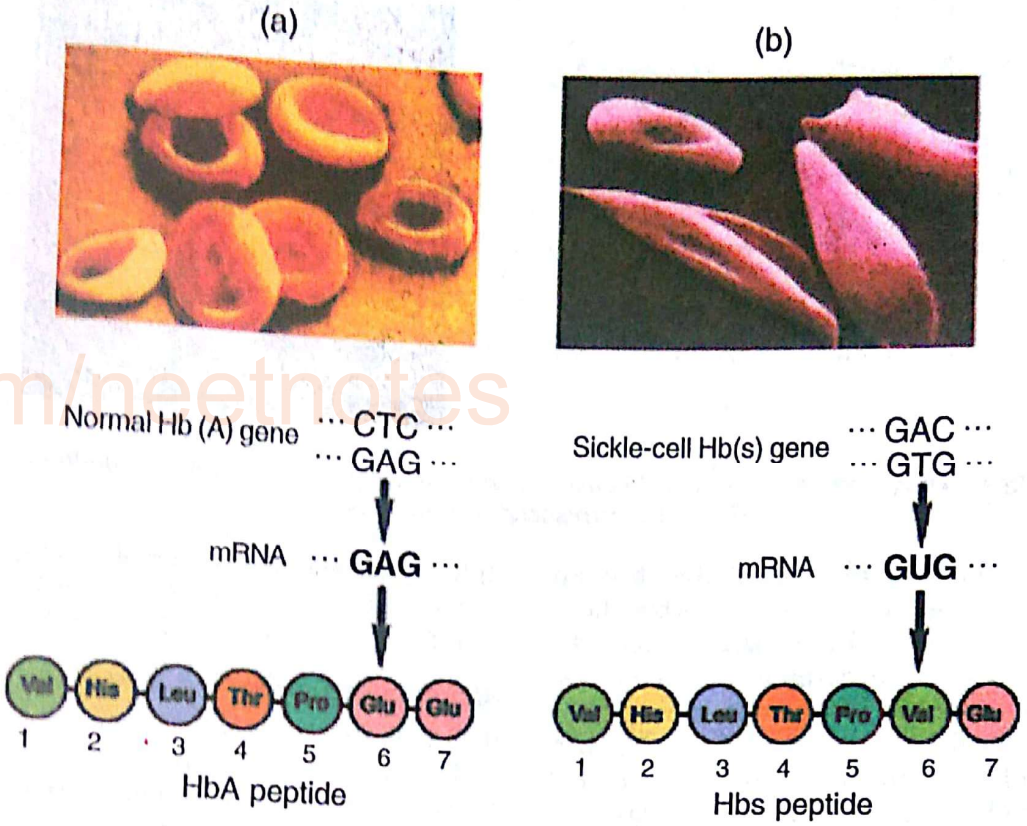

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Fig 13.15 *Micrograph of the red blood cells and the amino acid composition of the relevant portion of β-peptide of haemoglobin (a) From a normal individual (b) From an individual with sickle-cell anaemia*

sixth position in β chain of globin molecule of haemoglobin. The significance of this discovery has been not only to provide the direct evidence that genes specify proteins (see Chapter 14) but also to establish the concept of inherited molecular diseases.

Phenylketonuria

Phenylketonuria like alkaptonuria (see Chapter 14) is a case of inborn error of metabolism. It results in mental retardation and is inherited as an autosomal recessive trait. The affected individuals have a blocked metabolic step so that they are not able to convert the amino acid phenylalanine into amino acid tyrosine. This leads to accumulation of phenylalanine and its conversion to phenylpyruvic acid and other derivatives. Also, Phenylalanine and its derivatives accumulate in cerebrospinal fluid leading to mental retardation, and also get excreted in urine due to poor absorption by kidney.

Down's Syndrome

The previous two examples of genetic disorders are due to the mutant allele and their defective products. However, disorders can also be created by imbalance in chromosome number and chromosome rearrangement. One classical example of this category is the Down syndrome, first described by Langdon Down in 1866. The affected individuals have a very different but characteristic external appearance. They display prominent folding at the corner of eyes and have short statures. They have small round heads; protruding, furrowed tongues that cause the mouth to remain partially open; and short, broad hands with fingers showing characteristic fingerprint patterns. Physical, psychomotor, and mental development is retarded and the life expectancy is shortened. This condition arises due to an additional chromosome which has been identified as

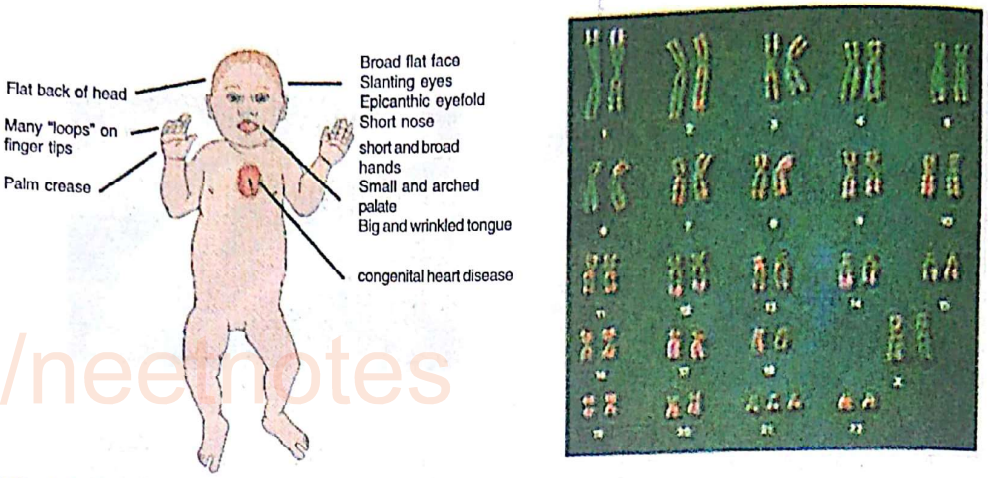

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Fig 13.16 *A representative figure showing an individual inflicted with Down's syndrome and the corresponding Karyotype*

chromosome 21. You may recall that human somatic cells contain 46 chromosomes but those with Down's syndrome will have 47 chromosomes, with one additional chromosome constituting three copies of chromosome 21 instead of normal two (Fig 13.16). Such a condition of chromosome addition is called trisomy which arises due to the formation of n+1 male or female gamete and the subsequent fertilization by a normal n gamete [(n+(n+1)]. The n+1 gamete is formed by the failure of paired homologues to separate at anaphase, a process known as **non-disjunction**. The higher age (35-40 years) of mothers has been implicated in producing such gametes with extra chromosome. Since this disorder is produced due to rare meiotic abnormality, it is not expected to run in families. However, in rare cases, it does so. Such instances are known as familial Down's syndrome and arise due to translocation of the major portion of chromosome 21 to chromosome 14. Translocations are a type of aberration we have already discussed in this Chapter. Such individuals will have a total of 46 chromosomes but partial trisomy of chromosome 21. They will exhibit Down's syndrome.

Alzheimer's Disease

It is a case of loss of memory and ability of judgement as well as general physical impairment. It results from accumulation of amyloid protein plaques in the brain resulting in the degeneration of neurons. The protein involved, amyloid – β peptide is produced and processed in a number of ways in the normal brain from a large amyloid precussor protein. This disease is common among Down syndrome (due to involvement of chromosome-21 trisomy). Different genes have been linked to Alzheimer's disease but these genes only predict susceptibility to disease.

Sex Chromosome-related Genetic Disorders

In humans, where each diploid cell contains 46 chromosomes, conditions such as 2A+XXY (47) or higher doses of X or Y or both lead to aberrant sexual development (Table 13.2).

Table 13.2. Aberrant Sexual Development

Chromosome composition	Chromosome number	Type of effect	Sexual differentiation
2A + XXY/XXXY	47/48	Klinefelter syndrome	Male
2A + XXX/XXXX	47/48	Female	Female
2A + XYY	47	Male	Male
2A + XO	45	Turner Syndrome	Female

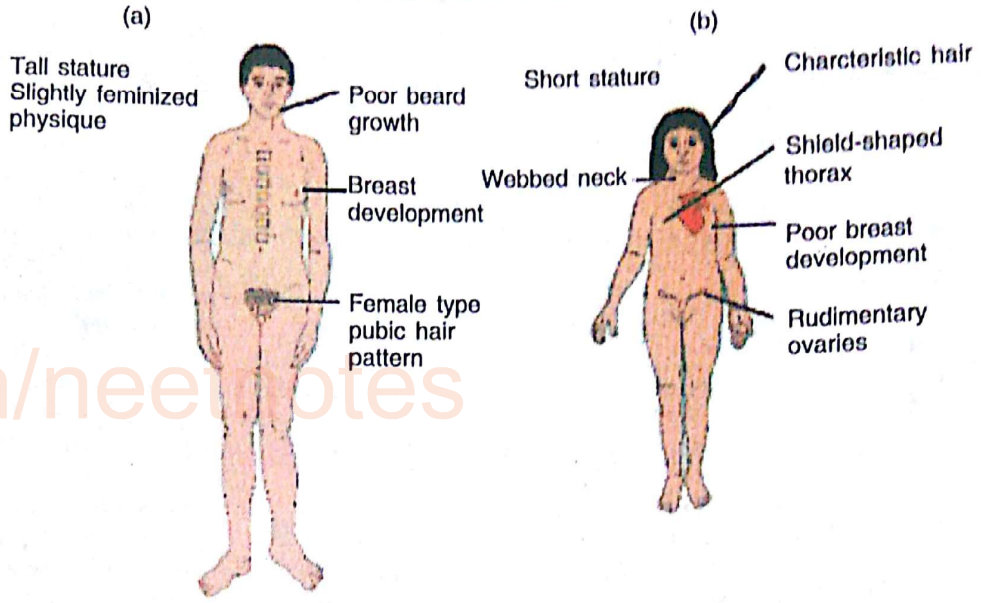

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Fig 13.17 *The associated phenotype of genetic disorders created by the imbalances in the sex chromosome composition in humans. (a) Klinefelter Syndrome, (b) Turner Syndrome*

The individuals with Klinefelter's syndrome, though have overall masculine development, feminine development is not completely suppressed, and are sterile (Fig 13.17a). In 2A+XXX condition (47), the females have usually normal genitalia, limited fertility, and slight mental retardation. Higher doses of X may lead to more pronounced effects. The addition of extra Y leads to 2A+XYY (47) condition. The males possessing such a condition show above-average height and subnormal intelligence. They are prone to psychopathic tendencies. Similarly, individuals having a single X chromosome 2A+X0 (45) have female sexual differentiation but ovaries are rudimentary. Other associated phenotypes of this condition (Turner's syndrome) are short stature, webbed-neck, broad chest, lack of secondary sexual characteristics and sterility (Fig 13.17b). Thus, any imbalance in the copies of the sex chromosomes may disrupt the genetic information necessary for normal sexual development.

SUMMARY

Discovery of chromosomes in the late 1800s and studies on their behaviour during nuclear divisions were instrumental in the rebirth of Mendel's work. The link between the unit factors (genes) and that of chromosomes during meiosis was reflected in the chromosome theory of inheritance. Various lines of studies were pursued to reveal this link. In one approach, chromosome structure in both, prokaryotes and eukaryotes was worked out and it was established that in both cases, it consists of Deoxyribose Nucleic Acid (DNA) and the associated proteins, but eukaryotes exhibit a higher degree of organisation.

In another line of study, location of genes on the chromosome was investigated. The alleles located on the same homologue were said to be linked as they may be usually transmitted together during gamete formation. However, the process of crossing over during meiosis results in the reshuffling of genes between the homologous chromosomes creating recombinants, thereby contributing to the genetic variability within gametes. Early in the last century, geneticists realised that crossing over could provide a basis of mapping the

linked genes relative to one another. This led to the linkage maps in a whole variety of organisms. Recombination also brings about the creation of new gene combinations, thus generating variability.

Investigations into the uniqueness of each organism's chromosomal constitution have further enhanced our knowledge of genetics. Firstly, in many sexually differentiated species, sex chromosomes have been identified. In such cases, all other chromosomes are referred to as autosomes. Genes located on the sex chromosomes follow a very characteristic pattern of inheritance. In humans, the individuals with altered sex chromosome compositions established that Y chromosome is responsible for male differentiation, while in *Drosophila* the same is controlled by the ratio between the number of X-chromosomes and the set of autosomes.

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While recombination is one important source of genetic variability, the most critical mechanism is mutation. Mutations arise due to alterations in the genes that may lead to changed function and thus a different phenotype. Mutation is the primary source of allelic variation.

Besides mutation, chromosomal changes involving number and structure can also lead to a lot of variations. In polyploidy, complete set of chromosomes are added but in aneuploidy there can be a gain or loss of individual chromosome(s). Large segments of chromosomes can be modified by deletions or duplications, bringing a change in gene number. Inversions and translocations alter the gene order along chromosome without making any change in the number of genes. Their heterozygosity may lead to abnormal meiosis and thus sterility.

EXERCISES

1. How would you correlate the behaviour of chromosomes at meiosis to
   (i) segregation of an allele pair
   (ii) independent assortment of two genes?
2. Differentiate between:
   (i) Complete linkage and incomplete linkage
   (ii) Crossing over and cross over gametes.
3. In a test cross AaBb x aabb, 90 per cent of the progeny are like parents. Determine:
   (i) the progeny type for the rest of the population?
   (ii) Are the genes linked?
   (iii) Is there any crossing over between the genes?
4. If the distance between the genes on a chromosome is as follows, prepare a genetic map, assigning the correct order of genes.
   a ——— b = 5cM
   b ——— c = 3cM
   a ——— c = 2cM
5. What will happen:
   (i) When complete sets of chromosomes are added to diploid genome?
   (ii) When individual chromosomes are added to or deleted from the diploid genome?
   (iii) When a part of the chromosome is lost?

CHAPTER 14

NATURE OF GENE : ITS EXPRESSION AND REGULATION

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By now, you know that parents transmit the heritable characters to the offspring. This forms the basis of maintaining a sort of continuity amongst the members of a species. Also, factors or genes responsible for these characters are located on the chromosomes, which through the gametes enter a diploid zygote cell composition that explains the process of transmission. This process is common to all sexually reproducing organisms. Thus, each offspring receives 50 per cent component of chromosomes and thus the genes, from male parent and 50 per cent component from female parent. In this Chapter, you will study more about genes, their structure and how their expressions are regulated.

By the time these facts were discovered, the cell was getting better understood in biochemical terms. This led to one important question as to which biomolecule contributes to the chemical form of gene, and whether structure of that biomolecule can explain all the functions assigned to a gene. On the basis of the genetic knowledge, a gene is expected to have following features:

(i) a gene should be able to store and express information to confer the heritable characters of the living organisms;

(ii) a gene should have the capability to make its own replica so that a copy can be transmitted to the offspring;

(iii) a gene should also have a mechanism to undergo mutations that will generate the biological variation so essential for evolution.

If we consider the major biomolecules of a cell and correlate their variety with the variety expected amongst different genes, proteins will attract our attention. While we expect each living organism to have different genes for corresponding phenotypes, we also have a whole variety of proteins formed. For a very long time thus, protein remained the strongest candidate as the chemical form of gene, even in the absence of any direct experimental proof.

14.1 NATURE OF GENETIC MATERIAL

While such experimental proofs were being sought, eukaryotic nuclei from a number of sources containing another biomolecule, deoxyribose nucleic acid or DNA, first described by Friedrich Miescher in 1868, became known. The role of cell nucleus was already known in hereditary process. Such information led to a fresh search for unravelling the biochemical nature of gene. But the answer came from an unexpected source.

In 1928, Frederick Griffith, a British scientist working on the pathogenicity of a bacterial strain, *Diplococcus pneumoniae* now called *streptococcus*, discovered the process of transformation. This strain causes bacterial pneumonia in mammals including humans. The disease-causing or *S*-strain has cells surrounded by a capsule, and forms smooth glistening colonies when grown on agar medium. Some mutant strains form rough colonies or *R* strains. This strain does not cause pneumonia. Also, when *S* strains are heat-killed and injected into mouse, no disease symptom appears. Surprisingly, however, when a mixture of heat-killed *S* type (inactivated) and live *R* bacteria (nonpathogenic) were mixed and then injected, disease reappeared. Live *S* type cells could be recovered from the blood of dead mice. Griffith proposed that a 'transforming principle', a chemical substance was released by the killed *S* cells which transformed the *R* bacteria into *S* type. This was a permanent genetic change as *S* type bacteria continued to produce similar cells (Fig. 14.1).

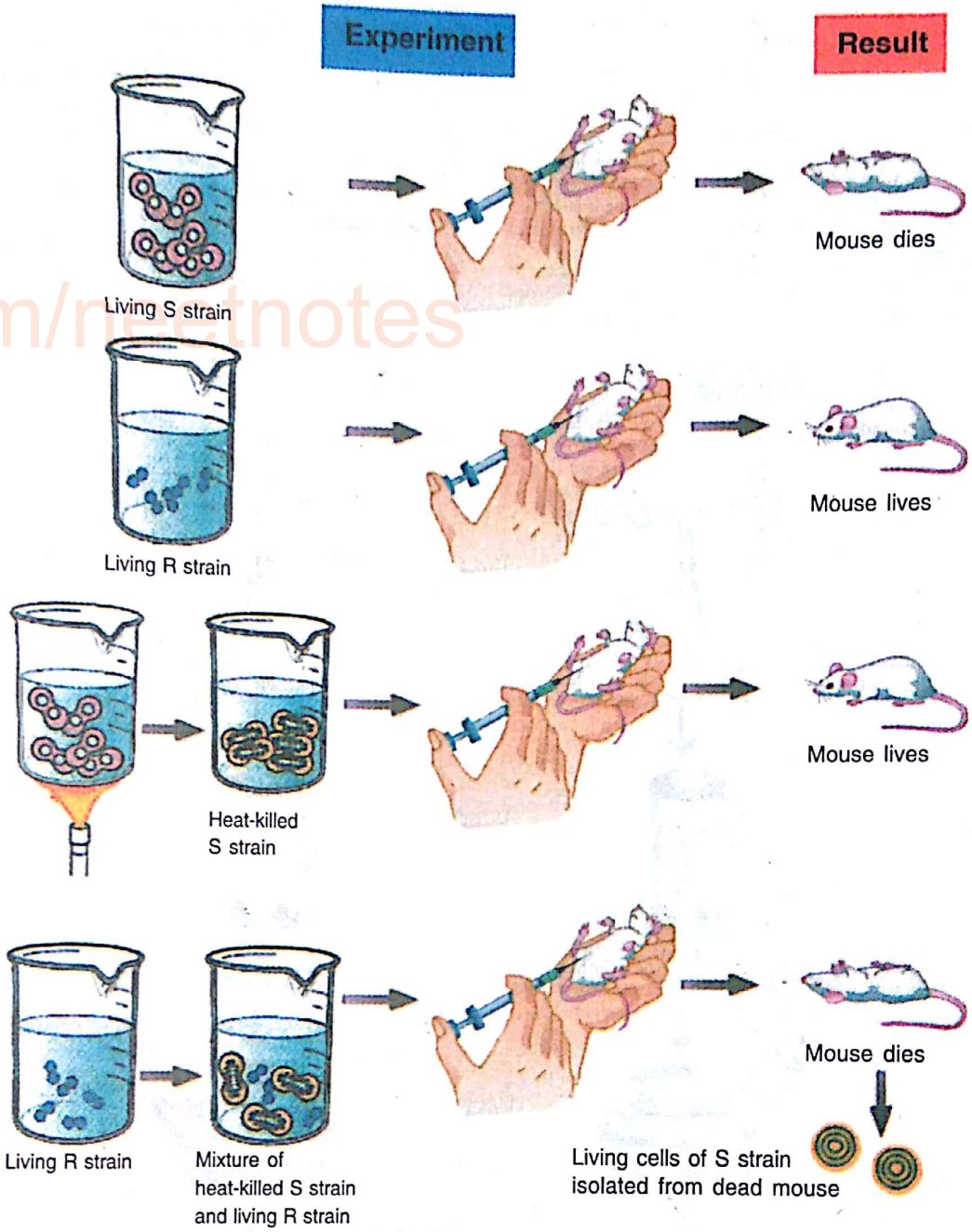

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Fig 14.1 *Griffith's experiment demonstrating transformation in Diplococcus bacterium*

In 1944, Oswald T Avery, Colin MacLeod and Maclyn McCarthy revealed the chemical nature of the transforming substance to be DNA. They showed that DNA isolated from S bacteria could by itself confer the pathogenic properties to *R* cells. This fact suggested that DNA has the genetic properties.

Another definitive evidence came from experiments on T_2 bacteriophage by Alfred D. Hershey and Martha Chase in 1952. A bacteriophage is simple in molecular constitution as it consists of a capsid that is made up of protein, and DNA that is contained within the head portion of the capsid. Hershey and Chase

based their experiment on the fact that DNA but not the proteins contains phosphorus, and similarly sulfur is present in proteins but not in DNA. They then incorporated radioactive isotope of phosphorus (^{32}P) into phage DNA and that of sulfur (^{35}S) into proteins of separate phage cultures. These phage types were used independently to infect the bacterium *Escherichia coli*. After sometime, this mixture was agitated on a blender to separate the empty phage capsids or 'ghosts' of the bacterial cells and the two were separated by centrifugation. Hershey and Chase showed that when ^{32}P was used, all radioactivity was associated with bacterial cells and if followed, appeared in the progeny phage. However, when ^{35}S was used, all radioactive material was limited to phage 'ghosts'. The conclusion was simple and straightforward: DNA alone is able to impart all the characteristics to the phage progeny and proteins did not play any role in this process (Fig. 14.2).

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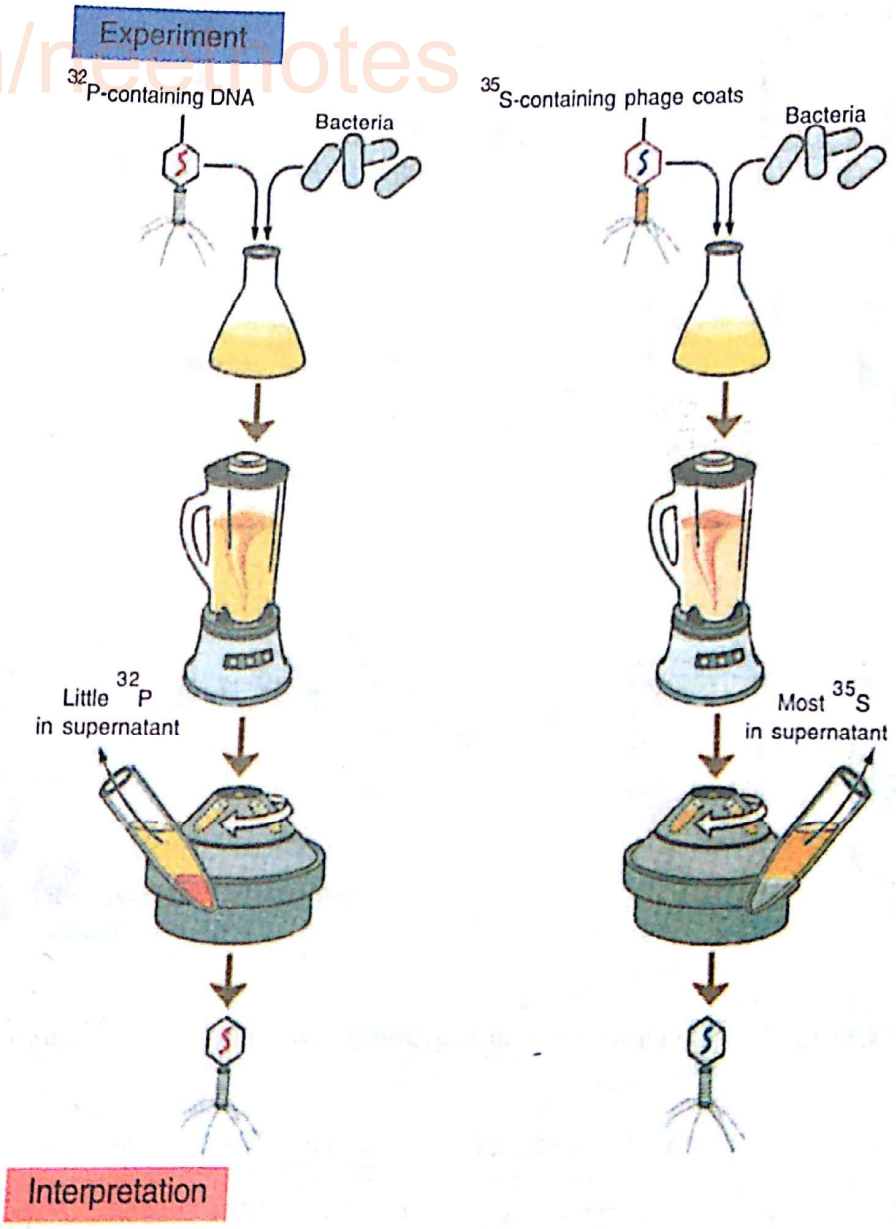

Fig 14.2 *Hershey and Chase's experiment demonstrating that only phage DNA injected in the host cell determines all the characteristics of the progeny phage*

With these two evidences and information linking DNA and nuclei and transmission through gametes involving principally nuclear material led to a fresh look at the structure of DNA with special reference to its function as genetic material.

14.2 DNA AND ITS STRUCTURE

By the time DNA structure was proposed by James Watson and Francis Crick in 1953, the chemical composition of DNA was already known. The deoxyribose nucleic acid (DNA) consists of four types of basic units called nucleotides (See Chapters 2 and 10). Each nucleotide is made up of a pentose sugar (deoxyribose type), a phosphate group, and a nitrogenous base. A subunit composed of only sugar and the nitrogen base is known as **nucleoside**. The four nucleosides differ from each other in the type of the base which could be adenine (A), guanine (G), thymine (T), or cytosine (C). The Adenine and Guanine are the purines and Thymine and Cytosine are the pyrimidines. In chemical terms, each nucleotide is a deoxy-5 -monophosphate, for example dAMP, dGMP, dTMP, or dCMP. So, DNA is a polynucleotide. Erwin Chargaff in 1949 had shown from his studies on DNA derived from a number of sources that there are certain empirical rules regarding the composition of bases in DNA known as **Chargaff 's rules**. These are:

(i) Total amount of purine nucleotides always equals the total amount of pyrimidine nucleotides i.e.,

[A]+[G] = [T]+[C].

(ii) The proportion of A is equal to T and so also of G is equal to C,

but amount of [A]+[T] is not neeessarily equal to [G]+[C].

Therefore, [A]= [T]; [G] =[C]

but $\frac{[A]+[T]}{[G]+[C]}$ = varies with the organism

In 1953, Maurice H.F. Wilkins and Rosalind E. Franklin took X-ray diffraction pictures of crystalline DNA. They concluded that it is a long molecule consisting of two similar strands running in parallel and helical manner. The helix makes one complete spiral turn every 3.4 nm and has a diameter of 2 nm (Fig.14.3a). Watson and Crick worked on these clues in the light of the functions assigned to a gene and developed their famous model 'Double Helix'. In this model, they proposed that DNA consists of two strands, which are helically coiled. Each strand consists of a backbone, made up of alternating deoxyribose sugar and phosphate with phosphate joining the two sugars through a phosphodiester bond (phosphate group forming a bridge between – OH groups of two adjacent sugars). The bases are stacked inside and pair with the base of the opposite strand through hydrogen bonds. Keeping in mind Chargaff's rule, they proposed that the pairing will always be between A and T, and G and C. The two strand runs antiparallel, i.e., one in 5'→3' direction and the other 3'→5' direction (Fig 14.3b).

The specific pairing between purine and pyrimidine bases proposed by Watson and Crick not only fitted very well with the radius of DNA determined from X-ray diffraction pictures but also had necessary complementary 'lock and key' shape for efficient hydrogen bonding, which happens to be two between A and T (A=T) and three between G and C (G=C). The stacking of bases creates two types of grooves called major and minor grooves. Each turn accommodates 10 bases. For this work on DNA structure, Watson and Crick along with Wilkins received Nobel Prize (Medicine and Physiology) in 1962.

Initially, two different forms of right-handed DNA helix A and B were identified. Of these, B is more hydrated and most frequently found in living cells. Subsequently, an additional form of DNA was detected. This was a left-handed form and based on its zigzag-like backbone called *Z* DNA. The right-handed DNA can acquire a left-handed structure, at least over a short distance and temporarily, and through this process can influence the gene expression.

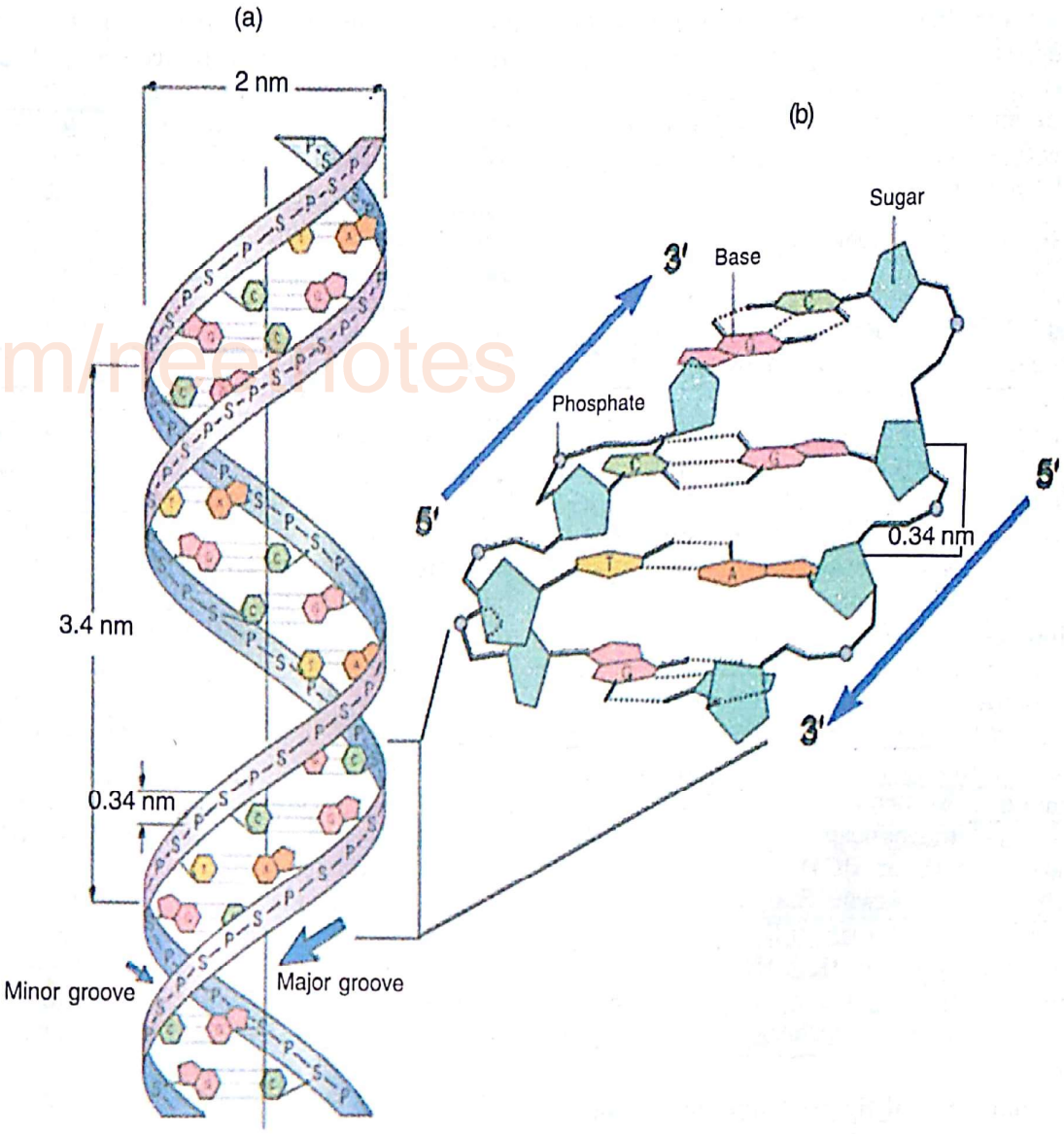

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Fig 14.3 *(a) General structure of a DNA double helix showing the position of sugar(S) phosphate(P) and bases, (b) The anti-parallel nature of the two strands. One strand starts with 5' and ends at 3' whereas the other beginning at 3' ends at 5'*

14.3 RNA AND ITS STRUCTURE

RNA or ribonucleic acid is another nucleic acid type. Like DNA, it is a polynucleotide but a number of differences are found in their structure. In RNA, the pentose sugar is ribose and not deoxyribose; also it contains uracil in place of thymine. Most RNAs are usually single stranded with partial double-stranded regions due to folding back of and pairing within its single chain. RNA serves as the genetic material in many viruses, some of which have double stranded RNA.

At least three major classes of cellular RNAs, all functioning during gene expression, are known. These are ribosomal RNA (rRNA), messenger RNA (mRNA), and transfer RNA (tRNA). All the three types of molecules originate as complementary copies of one of the two strands of a DNA segment that constitutes a gene, during the process of transcription. This would mean that RNA will have the same sequence as the other strand of DNA, except that uracil will replace thymine against adenine.

These RNA types can be differentiated on the basis of their size, sedimentation behaviour and genetic functions.

Ribosomal RNA is generally not only the largest but also the most prevalent of the cellular RNA species. This is an important structural component for protein synthesis. Messenger RNA, as the name suggests, carries the genetic message from DNA. Their length and sequence vary depending upon the gene which is being transcribed into mRNA. Transfer RNA, the smallest of the three types, carries amino acids to the ribosomes during translation. This species of RNA contains number of modified bases.

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14.4 DNA AND GENE

Now that we know the structure of DNA, we shall correlate this structure with the possible functions of a gene. Watson and Crick had believed that the specific pairing of the bases in the helix will play a key role in the transmission of the genetic character, both from one cell to its successors and also in gene expression.

DNA Replication

One of the prime functions of a gene is to make its copies, which could be transmitted to the daughter cells. This would help in maintaining the uniform genetic composition. Watson and Crick proposed that each DNA strand of a double helix can act as template for the synthesis of daughter strand in which bases are incorporated by specific hydrogen-bonded pairing (A with T, and G with C). The resulting DNA strand will be complementary to the template strand and identical to the other strand. As seen in the Fig.14.4a, the two daughter helices are exact replicas of the original double helix. The copying of DNA to make more DNA is known as DNA replication. In the process outlined above, since the daughter DNA consists of one old and one new strand, the mode of replication is called **semiconservative.**

The semiconservative DNA replication was confirmed by an elegant experiment conducted by M. Meselson and F.W. Stahl in 1958. They grew *E. coli* cells in the presence of heavy isotope ^{15}N (in the form of $^{15}NH_4Cl$) for several generations. This led to the incorporation of

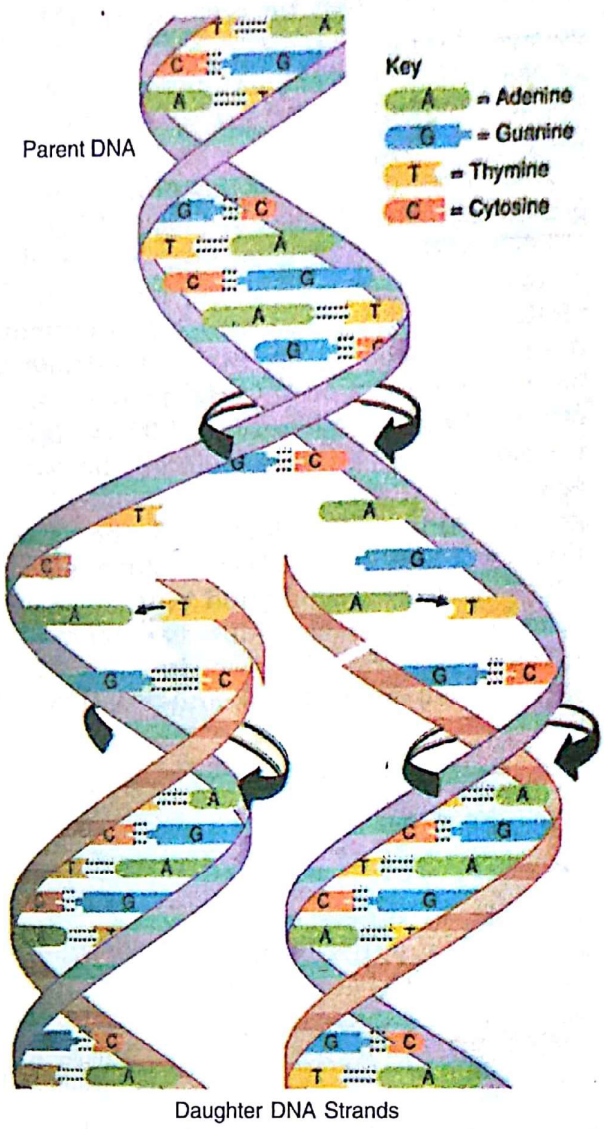

Fig 14.4 *(a) Semi-conservative mode of DNA replication. The two resulting helices are exact replica of the parentals duplex*

heavy isotope in all nitrogen-containing compounds including bases.

Such cells were then shifted to normal ^{14}N-nitrogen for one or two generations. DNA was isolated from the cells at each generation and the DNA molecules containing ^{15}N and ^{14}N were separated on CsCl-equilibrium density gradient centrifugation. When a solution of Cesium chloride (CsCl) is spun in a centrifuge at a very high speed (50,000 revolutions per minute) for many hours, the salt tends to settle down in the centrifuge tube creating a density gradient.In this gradient, the highest ion

concentration will be encountered at the bottom. When DNA is mixed with CsCl, it will finally settle at some point in the tube where the centrifugal force balances the buoyancy of DNA.

DNA containing heavy isotope ^{15}N has greater density and is called heavy DNA. Such a DNA settles at the heavier range of the gradient. After one generation, in ^{14}N medium, all DNA acquired an intermediate density whereas after two generations two bands were seen, one at intermediate and the other at 'light' DNA density. If we follow Fig14.4b, we can explain these results on the basis of semiconservative mode of replication. After one generation in ^{14}N medium, both the daughter DNAs will have $^{15}N/^{14}N$ composition instead of $^{15}N/^{15}N$ of the original and thus will have an intermediate density. After two generations, from a $^{15}N/^{14}N$ DNA two daughter helices formed will be $^{15}N/^{14}N$ and $^{14}N/^{14}N$. This explains the two corresponding bands. Just a year before Meselson and Stahl's experiment, Taylor had shown semiconservative mode of replication at chromosomal level in bean root tip cells.

Mechanism of DNA Replication

Though the mode of replication looks simple, the whole process is much complicated. A whole range of enzymes are required to take care of not only the various steps, and the structural and mechanistic constraints, but also to maintain the accuracy of the process. The DNA replication in most bacteria starts at a single point, **Origin of Replication** or *Ori*, and moves bidirectionally. In eukaryotes, there are several points of origin on the length of the DNA per chromosome. The first requirement before any synthesis can take place, is to unwind the double helix, so that the two stands are free to act as templates. This function is carried out by

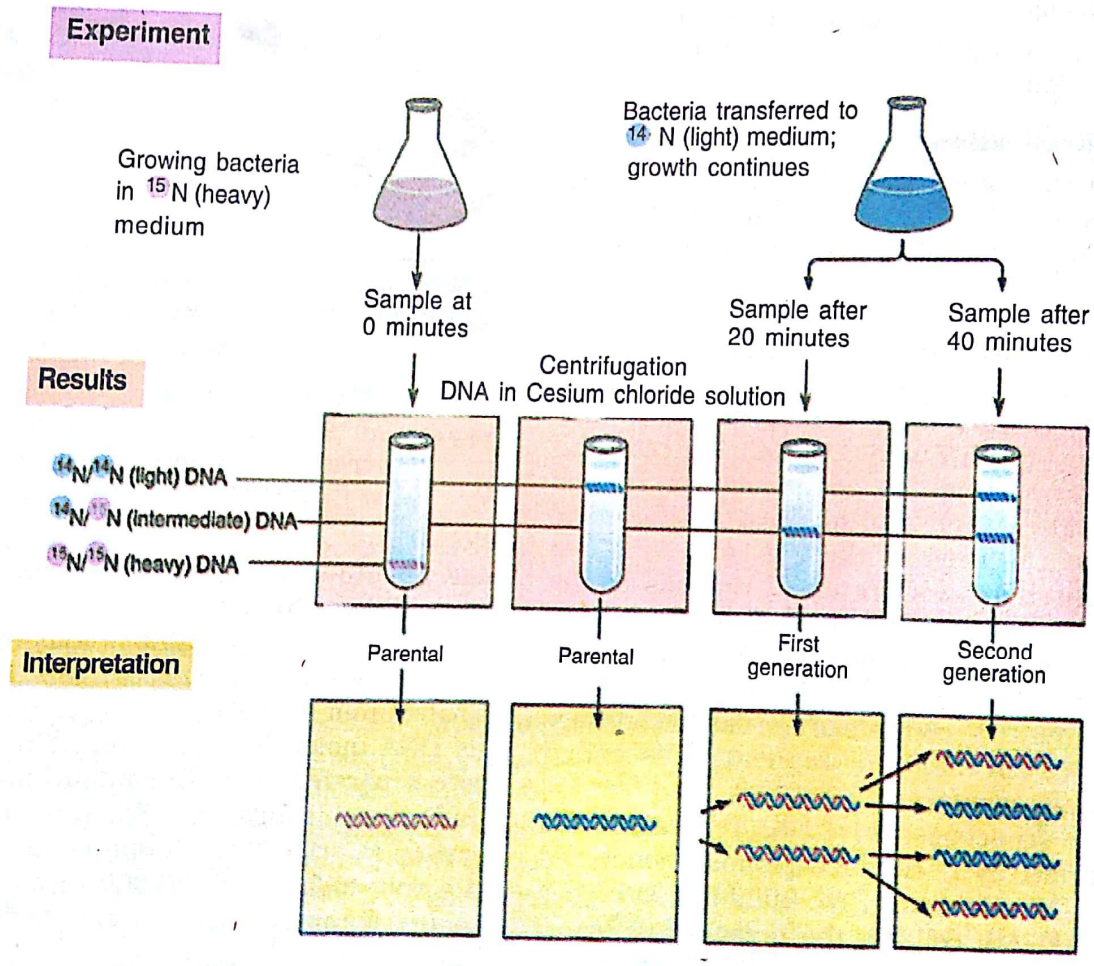

Fig 14.4 (b) Meselson and Stahl's experiment to prove the semi-conservative replication of DNA

the enzyme **Helicase,** which unzips the two strands beginning at the *Ori* site. As unwinding proceeds, other proteins called single-stranded binding proteins, associate with single strands and stabilise this condition. Unwinding also creates a coiling tension ahead of the moving **Replication fork**, a structure that will be formed when DNA replication begins. This tension is reduced by **Topoisomerases**. Figure 14.5 depicts these and the subsequent steps in DNA replication.

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The most important DNA synthesising enzyme is DNA polymerase III. It along with other DNA polymerases (I and II) has the capability to elongate an existing DNA strand but cannot initiate the synthesis. All the three DNA polymerases function in 5'→ 3' direction only for DNA polymerisation and have 3'→ 5' exonuclease activity. Thus, to initiate DNA synthesis, a small segment of RNA, called an RNA primer complementary to the template DNA is synthesised by an unique RNA polymerase known as primase. It is to this primer, that DNA polymerase III adds 5'-deoxyribonucleotides and extends the DNA (Fig. 14.5). A problem will be encountered, as we know that two strands of DNA run antiparallel to each other and DNA polymerase III can function only in 5' → 3' direction. This problem is solved in the following manner. While on the one strand, the DNA synthesis is continuous and in 5'→ 3' direction, on the other strand, DNA is synthesised in small stretches resulting in discontinuous DNA synthesis. This happens in the opposite direction to the first strand but maintains the overall 5'→ 3' direction as required. Such a process is also referred to as semi-discontinuous replication. The short stretches of DNA, each primed by RNA are called **Okazaki fragments**; named after the Japanese scientist who discovered them. RNA primers are then removed, and the gap is filled by DNA nucleotide. Both steps are performed by DNA polymerase I. These fragments are then sealed by the enzyme **Ligase**. The strand which supports the continuous DNA nucleotide is the **leading strand** and the one which is replicated in short stretches is called the **lagging strand**.

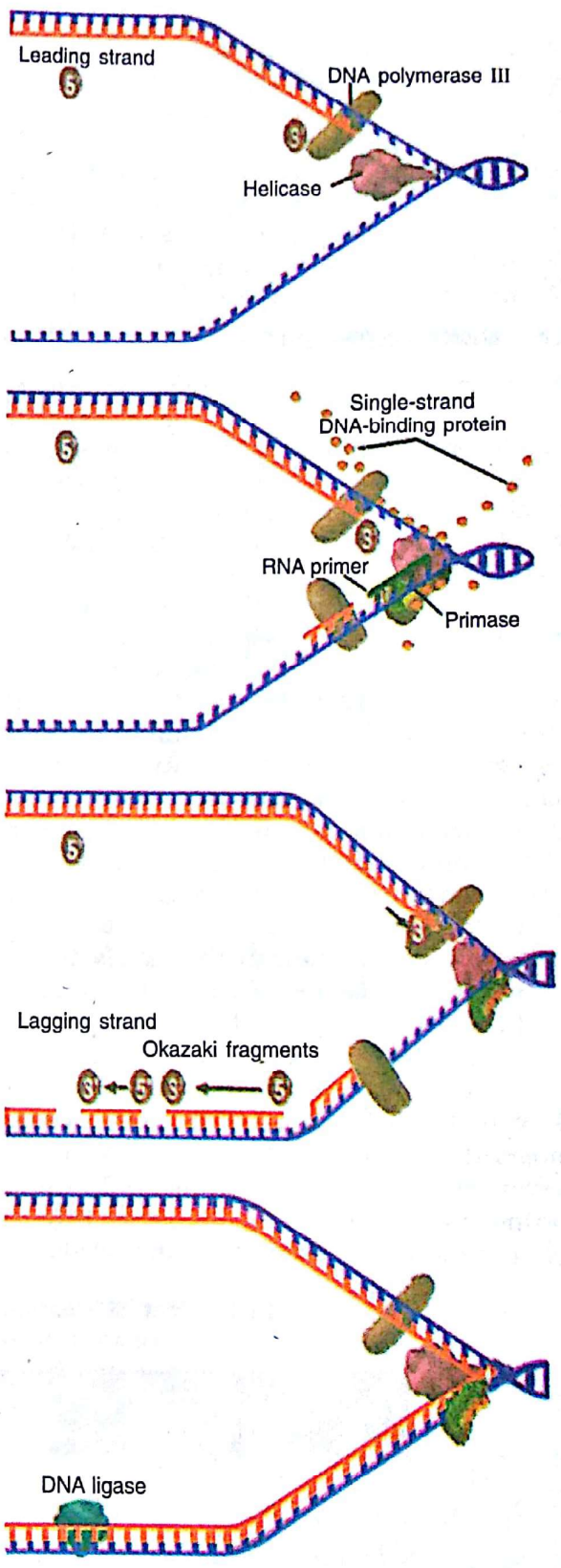

Fig 14.5 *The molecular mechanism of semi-discontinuous DNA replication*

The process of replication also ensures the accuracy so as to maintain the nucleotide sequence of the original DNA. Firstly, the DNA polymerase action itself is very accurate. If, however, a wrong nucleotide is inserted, both DNA polymerases I and III are able to detect and excise the wrong base. Even though DNA synthesis is slower in eukaryotes as larger DNAs need to be replicated, the general steps of replication are similar to those of prokaryotes.

14.5. GENE EXPRESSION

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The second important characteristic of the gene is to store and express the genetic information that will contribute towards the phenotype, and will be passed on to successive generations. In the following discussion, we shall explore as to how DNA can fit into this characteristic and how exactly it controls the gene expression.

The idea that genes control the metabolism was put forward as early as 1902 by Garrod. He studied several human disorders, which seemed to be inherited. He called them inborn errors of metabolism. In one such disorder, alkaptonuria, afflicted individuals cannot metabolise homogentisic acid, as a result it gets accumulated and is excreted in the urine. The oxidation products of this molecule are black and thus can be easily detected by the blackening of the urine upon exposure to air. The products also tend to accumulate in the cartilaginous areas causing darkening of ear and nose, and deposition in joints lead to mild arthritis. Interestingly, Garrod found that this disorder is inherited as a recessive trait and reasoned that normal individuals must be metabolising homogentisic acid. In affected individuals, however, the metabolic pathway is blocked resulting into alkaptonuria. Subsequently, a number of human diseases were identified and described which fell into the category of inherited metabolic defects. It thus led to the idea that many disorders may be inherited as any other gene and that genes function by controlling the metabolism.

The direct proof for this gene-metabolism relationship came from the work of George Beadle and Edward Tatum in the early 1940s on the fungus *Neurospora crassa*. They irradiated the spores with X-rays and isolated a range of nutritional mutants, which had requirement for an additional specific nutrient. Such mutants were called auxotrophs in comparison to the wild-type (prototroph) which could grow on a simple nutrient medium (minimal medium) containing a few salts and a sugar. For example, a number of arginine-requiring mutants were isolated which could be classified into three types:

(i) some could grow on ornithine-, or citrulline-, or arginine - containing medium,
(ii) some could grow on citrulline- or arginine-containing medium, and
(iii) some could grow only on arginine supplemented medium (Table 14.1).

It means that all mutants could grow on arginine-supplemented medium suggesting that arginine is the final product of this pathway, and since some could not grow on ornithine, this compound must be synthesised earlier than citrulline and/or arginine. Beadle and Tatum taking clue from this growth behaviour of *Neurospora* and other experiments proposed the arginine biosynthetic pathway (Fig.14.6).

The precursor compound first gives rise to ornithine, ornithine then gives rise to citrulline and the latter is finally converted to arginine; each of these steps is mediated by an enzyme. The mutants of class (i) could grow on ornithine, citrulline, or arginine suggesting that they lacked the capacity to synthesise ornithine but

Table 14.1 Effect of Medium Supplements on the Growth of Mutants of *Neurospora crassa*

Mutant types	Growth on medium supplemented with		
	Ornithine	Citrulline	Arginine
(i)	+	+	+
(ii)	-	+	+
(iii)	-	-	+

+ growth; - no growth

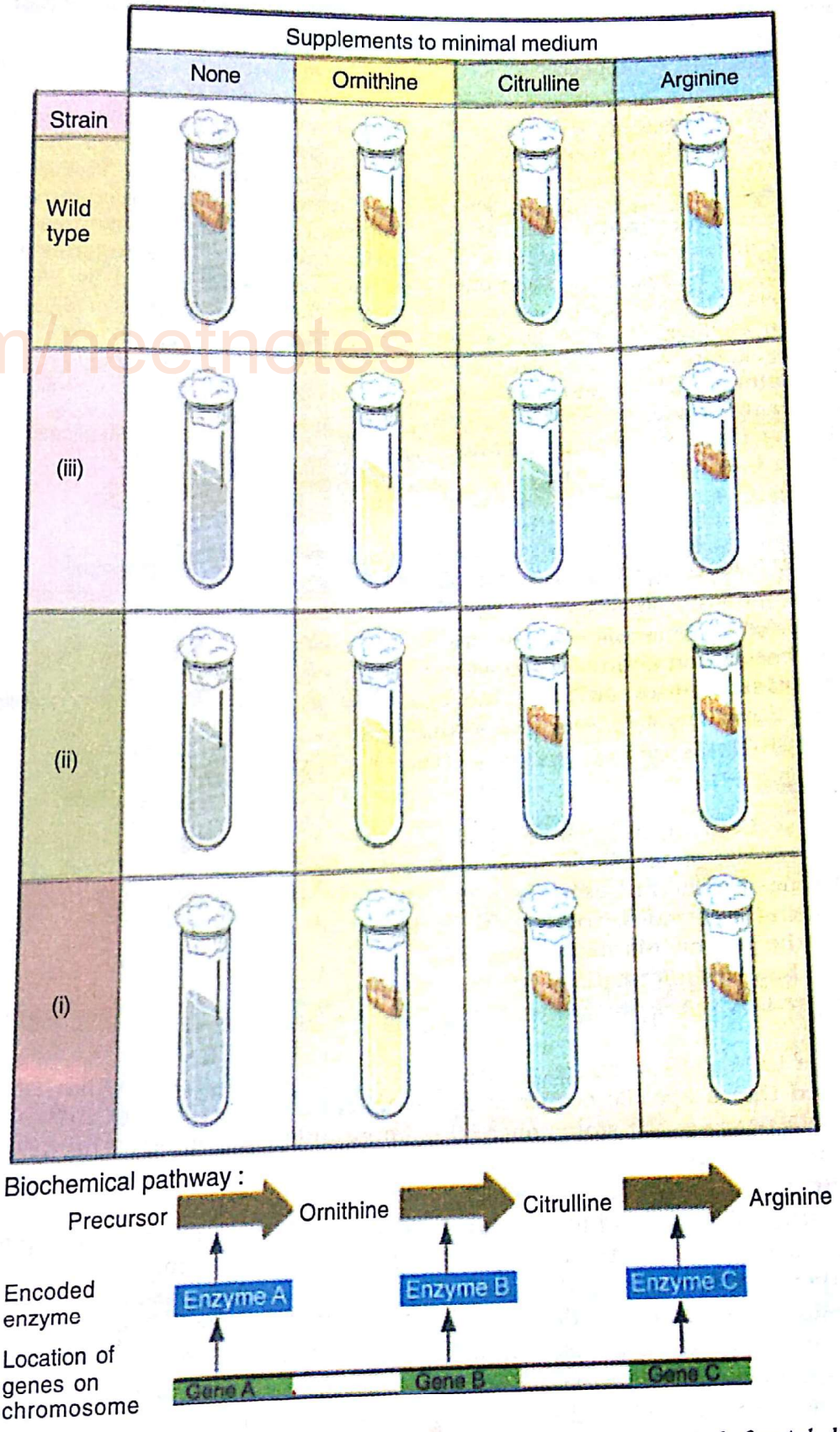

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Fig 14.6 *Beadle and Tatum's experiment on Neurospora crassa revealed the control of metabolism by genes*

beyond that they could complete the pathway. Similarly, mutants of class (ii) could not convert ornithine to citrulline but if supplied citrulline could synthesise arginine. The last class of mutant (iii) could not convert citrulline to arginine and had to be supplied the latter for growth. They reasoned that these defects could arise due to defective enzymes in each case. Since such changes were mutational, they opined that one gene controls one enzyme in a pathway leading to their famous one **gene-one enzyme** hypothesis.

Two ideas soon modified the one gene-one enzyme hypothesis. Firstly, while nearly all enzymes are proteins, all proteins are not enzymes. Also, many proteins are made up of subunits called polypeptides, with each distinct polypeptide under the control of a gene.

The adult human haemoglobin, for example, consists of 4 polypeptides-2α and 2β and a separate gene codes each type. This led to re-evaluation of Beadle and Tatum's hypothesis into **one gene-one polypeptide.** Further modification came when gene was identified as a functional unit or cistron and the same was called as **one cistron-one polypeptide.** Basically, all the three tenets suggest that the genes control the polypeptides.

Gene and Protein

In the previous section, we have established that DNA is the genetic material and we have also seen that genes function by exerting their control over proteins or enzymes. Now, we shall see as to how is the genetic information stored in DNA and then how this information coded in DNA is transmitted in the process of protein synthesis.

The work done in the late 1950s and early 1960s established that a specific sequence of 4 bases in the DNA serves as the store house of all genetic information or in essence is the basis of all life on Earth. The unique way in which these bases are arranged serve as what is known as genetic code, which determines the basic structure and therefore function of the whole variety of proteins. Thus, genetic code can be equated with Morse code used in telegraphic signal transmission. We shall describe in detail how gene expression occurs and how the genetic code was deciphered.

14.6 CENTRAL DOGMA OF MOLECULAR BIOLOGY

The expression of the genetic material occurs generally through the production of proteins. This involves two consecutive steps. These are **transcription** and **translation.** In transcription, the genetic information, stored in DNA, is transferred to an RNA, which in turn uses this information to direct the synthesis of proteins during translation. This unidirectional flow of information was described by F.H.C. Crick in 1958 as the 'Central dogma' of Molecular Biology (Fig. 14.7).

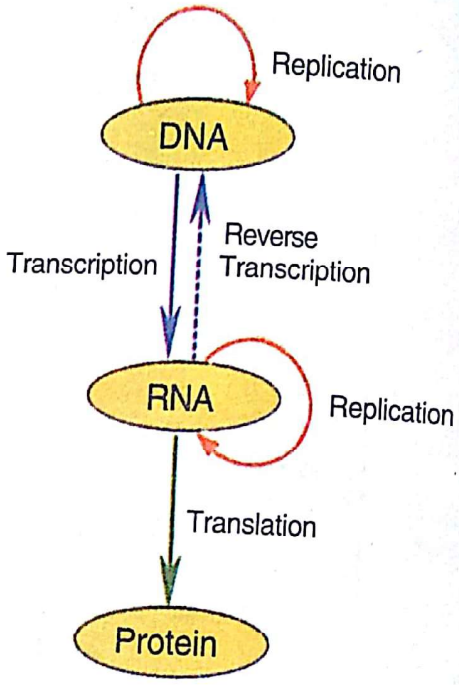

Fig 14.7 *The flow of genetic information*

You have already known that DNA is a self-replicating molecule. In 1970, however, an important modification of this information flow was brought to light by the work of H.M. Temin and D. Baltimore. Many tumor viruses contain RNA as genetic material and replicate by first synthesising a complementary DNA. This process is called **reverse transcription**. It is carried out by an RNA-dependent DNA polymerase called reverse transcriptase. These viruses are known as retroviruses and include Human Immunodeficiency Virus (HIV) that causes AIDS.

How the coded information of DNA is decoded into proteins? We will now describe the two major steps of the process.

Transcription

As described earlier, transcription generates a single-stranded RNA identical in sequence with one of the strands of the DNA. All the three species of RNA are produced through transcription. The strand of DNA that directs the synthesis of the mRNA via complementary base pairing is called coding or sense strand and acts as template for mRNA synthesis. The other strand is known as the non-coding strand or antisense strand. Transcription is accomplished by an enzyme **RNA polymerase** that gets physically associated with DNA. Only one type of such an enzyme is found in prokaryotes in contrast to eukaryotes where three different forms of RNA polymerase are found. RNA polymerase I, II and III catalyse the synthesis of rRNA, mRNA and tRNA, respectively.

RNA polymerase binds to a region of DNA called promoter which is recognised by sigma (σ) subunit of the enzyme RNA polymerase in prokaryotes and many transcription factors in eukaryotes. RNA polymerase not only initiates but also extends the RNA (chain elongation) and functions always in 5' to 3' direction. The process terminates once the enzyme completes the transcription of entire DNA segment and a termination sequence is reached (Fig. 14.8).

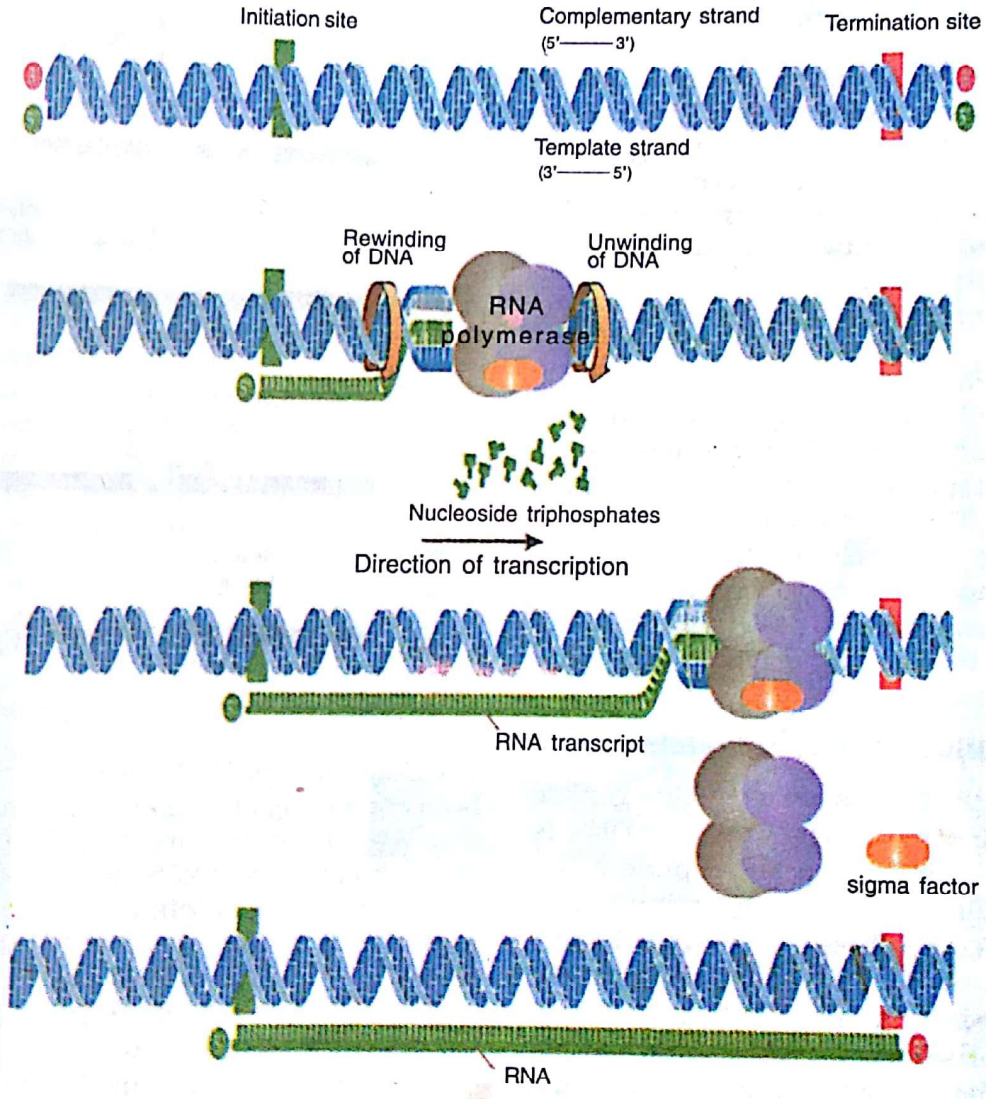

Fig 14.8 Transcription in prokaryotes-Enzymatic synthesis of RNA by RNA polymerase

In many bacteria, genes of related functions are grouped together and are called operons. An operon acts as a single transcription unit and thus produces several mRNA strands or polycistronic mRNA. In eukaryotes, only monocistronic mRNAs are generally produced.

Unlike the situation in prokaryotic genes, transcription in eukaryotes occurs within the nucleus and mRNA moves out of the nucleus into the cytoplasm for translation. The initiation and regulation of transcription is more extensive than prokaryotes. Another major difference between prokaryotes and eukaryotes lies in the fact that in eukaryotes the primary RNA transcript is further processed to form mRNA, a process called maturation. Initially at the 5' end a cap (consisting of 7-methyl guanosine or 7 mG) and a tail of poly A at the 3' end are present (Fig.14.9). The cap is a chemically modified molecule of guanosine triphosphate (GTP). The primary eukaryotic mRNA transcript is much longer and localised in the nucleus, when it is also called heterogenous nuclear RNA (hnRNA) or pre-mRNA. The eukaryotic primary mRNAs are made up of two types of segments: non-coding introns and the coding exons. The introns are removed by a process called RNA splicing. Of a pair of small nuclear ribonucleoprotein (SnRNPs pronounced "snurps"), one binds to 5' splice site and the other to 3' splice site. A spliceosome forms because of interaction between SnRNPs and other proteins. This spliceosome uses energy of ATP to cut the RNA, releases the introns and joins two adjacent exons to produce mature mRNA. Besides, these two post-transcriptional modifications, RNA editing may also take place before translation begins.

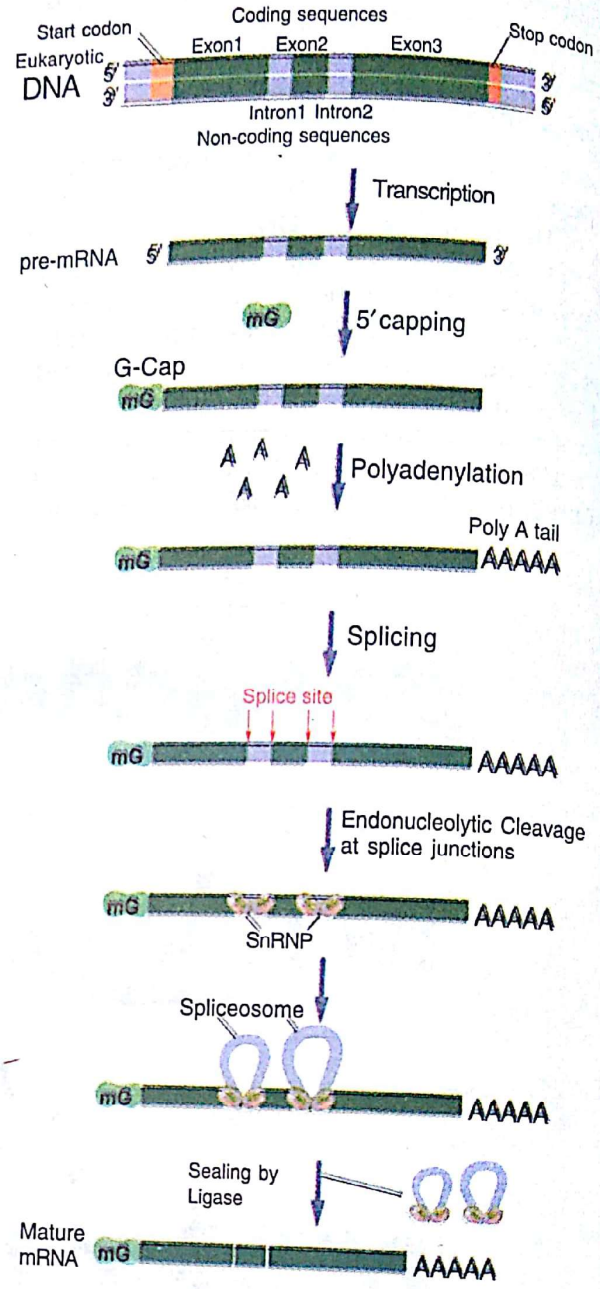

Fig 14.9 Transcription in eukaryotes

Translation : Biosynthesis of Proteins

Translation is the process in which the genetic message carried by mRNA from the DNA is converted in the form of a polypeptide chain having a specific sequence of amino acids. Besides mRNA, the three key players in this process are: ribosomes, tRNA, and amino acids. Ribosomes are the ribonucleoprotein particles that provide the sites for protein synthesis. On the basis of their sedimentation coefficients, prokaryotic and eukaryotic ribosomes are characterised as 70S and 80S, respectively, both consisting of a small and a large subunit (Fig. 14.10). In prokaryotes, the large or 50S subunit consists of 23S rRNA and a 5S rRNA + 32 different proteins and the 30S small subunit is made up of 16S rRNA and 21 different proteins. The eukaryotic large subunit (60S), on the other hand, is composed of 28S rRNA, 5S rRNA and 5.8S rRNA and 50 proteins and the 40S small subunit consists of 18S rRNA and 33 different proteins .

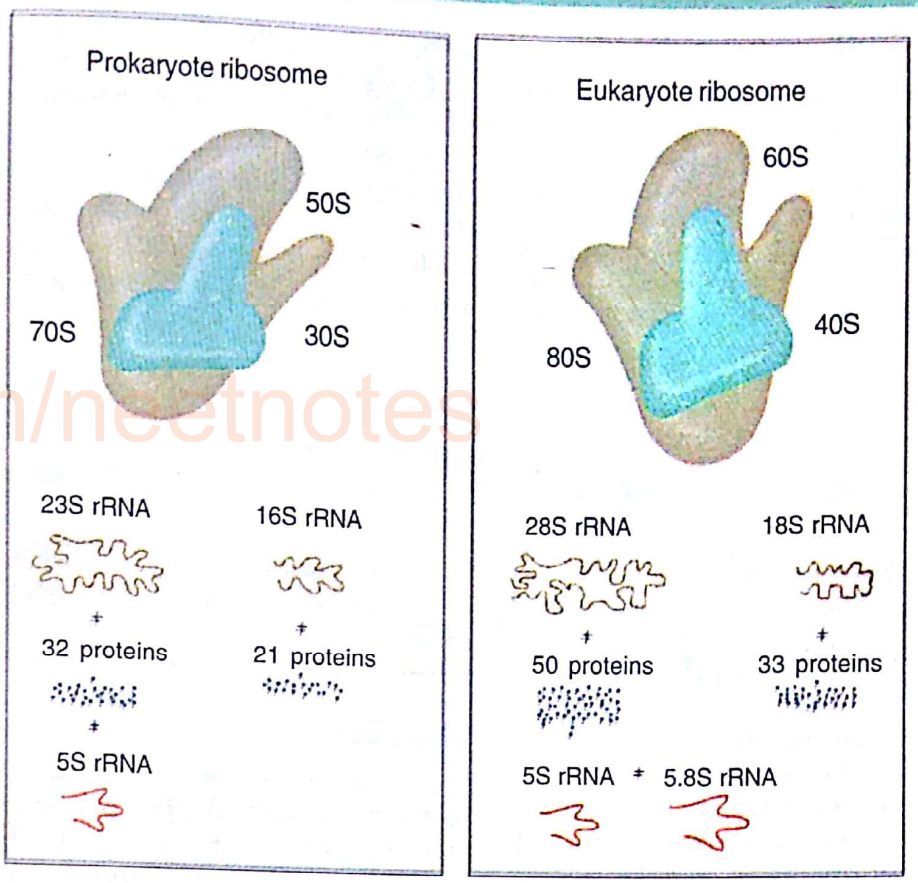

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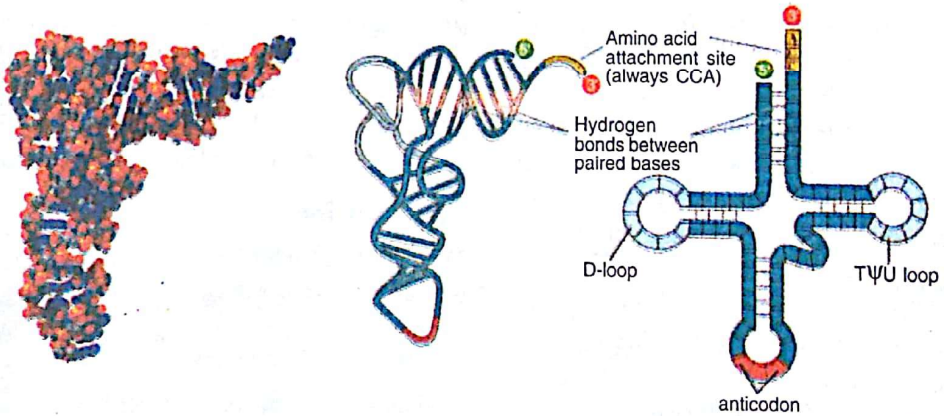
Fig 14.10 *Generalised structure of ribosome in prokaryotes and eukaryotes*

The tRNA, smallest of the three RNA species known displays nearly identical structure in pro- and eukaryotes. In 1965, R.W. Holley proposed a clover-leaf model of tRNA which results from intra-strand base pairing creating paired stems and unpaired loops. The 3' end of tRNA contains the sequence for amino acid attachment. The anticodon loop carries an anticodon complementary to the known amino acid code for which it is specific. In three dimensions, tRNA looks like an L-shaped molecule (Fig.14.11).



3D- computer generated space-filling structure of a t RNA

3D- L shaped structure of a t RNA

2D- clover leaf model of a t RNA

Fig 14.11 *Structure of tRNA*

Steps in Translation

Twenty naturally occurring amino acids participate in protein synthesis- Translation demands that a tRNA is specifically linked to an amino acid, a process known as charging. This is brought about under the direction of an enzyme, aminoacyl tRNA synthetase which is highly specific, i.e. each recognizes only one amino acid. Charging is a two-step process:

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Step 1

Amino acid* + ATP $\xrightarrow{\text{Aminoacyl tRNA Synthetase*}}$ **Aminoacyl adenylic acid + P-P**

Step 2

Aminoacyl adenylic acid + tRNA* $\longrightarrow$ **Charged tRNA (tRNA - Amino acid*) = P-A + aminoacyl tRNA synthetase**

* *Represents specific reaction between these components.*

In the first step an amino acid reacts with ATP in the presence of a specific aminoacyl tRNA synthetase to produce the activated form or an aminoacyl adenylic acid. While still associated with the enzyme, the complex now reacts with a specific tRNA and the amino acid is transferred to the adenine of 3' end of tRNA, and the enzyme leaves the complex.

The process of translation involves three steps, *viz.* initiation, elongation, and termination (Fig.14.12).

Initiation

The translation of mRNA begins with the formation of initiation complex. For this, small subunit of ribosome, a mRNA, a specifically charged initiator tRNA, GTP, Mg^{2+} and proteinaceous initiation factors are assembled. These initiation factors are designated as IFs in prokaryotes and eIFs in eukaryotes. The initiation factors bind to the smaller ribosome subunit and then this complex binds to a sequence of mRNA, preceding the initiator AUG codon. The initiator AUG in prokaryotes codes for formylmethionine (formylmet) but in eukaryotes, this code is for methionine.

Binding of charged formyl-met tRNA to all the components assembled at 30S subunit forms the initiation complex. Later, when the large 50S subunit joins the initiation complex, the initiation factors are released and complete 70S ribosome is formed. The ribosome has binding sites for two charged tRNAs labelled as P (peptidyl site), and A (aminoacyl site). The initiator tRNA is positioned at P site leaving the triplet (three bases of mRNA or codon) at the A site unoccupied to allow the entry of another charged tRNA (Fig. 14.12).

Elongation

Ribosome moves along the mRNA in the 5' to 3' direction. A charged tRNA whose anticodon is complementary to the second codon enters the empty A site. Once this happens, the peptidyl transferase, links the two amino acids through a peptide bond. The 23S rRNA of large subunit has been assigned this catalytic function. The initiator tRNA now delinks, and the dipeptide so formed is translocated to the P site. The movement of entire mRNA - tRNA dipeptide shifts by a codon exposing the next codon at A site in a process called translocation. All these steps are assisted by a number of proteins called **elongation factors (EFs)** and the energy from the hydrolysis of GTP (Fig. 14.12). In prokaryotes, elongation factors are called EF-Tu, EF-Ts and EF-G. Eukaryotes require a more complex set of accessory factors.

Termination

The sequence of elongation continues till the whole of mRNA is translated and a signal in the form of termination codon (UAG, UAA, or UGA) is reached. These do not code for any amino acid and, therefore, the translation stops. They also signal a GTP-dependent release factor (RF_1, RF_2 and RF_3) which cleaves the polypeptide from the terminal tRNA, releasing it from the translation complex (Fig. 14.12).

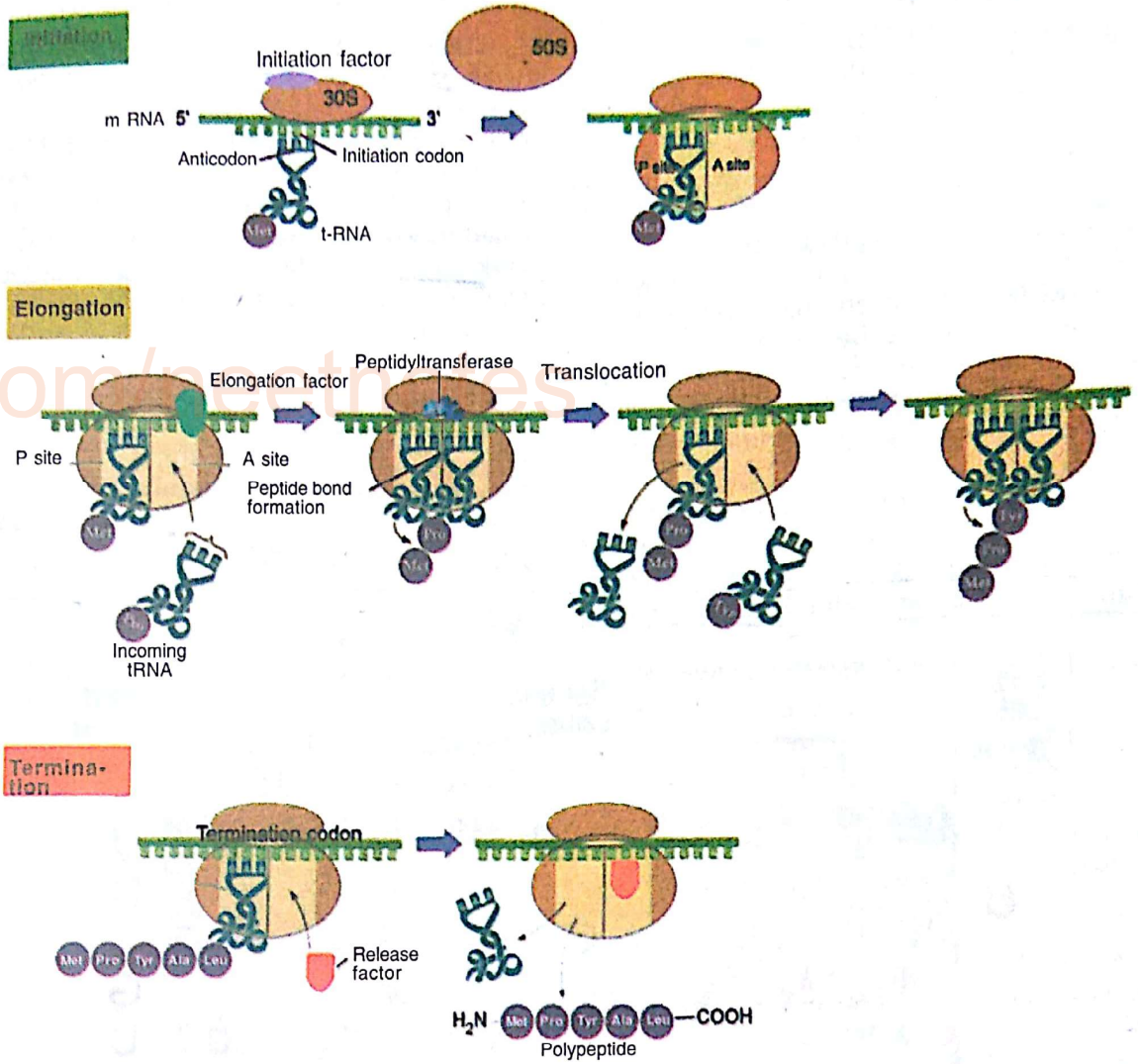

Fig 14.12 *Stages in translation – Initiation, elongation and termination. The net result is the synthesis of a peptide in which amino acids are aligned according to the message received from the gene*

The polypeptide formed under the guidance of the mRNA thus consists of a specific sequence of amino acids. Once the initial portion of mRNA is translated, it is free to engage into another round of translation. This process can be repeated several times creating a structure called polyribosomes or polysomes.

Some antibiotics inhibit bacterial protein synthesis. This forms the basis of inhibiting the bacterial invaders and stemming the infection without harming the human host (Table 14.2).

Table 14.2 Some Inhibitors of Bacterial Protein Synthesis

Antibiotic	Effect
Tetracycline	inhibits binding of amino-acyl tRNA to ribosome
Streptomycin	inhibits initiation of translation and causes misreading
Chloramphenicol	inhibits peptidyl transferase and so formation of peptide bonds
Erythromycin	inhibits translocation of mRNA along ribosome
Neomycin	inhibits interaction between tRNA and mRNA

14.7 GENETIC CODE

The process by which the information coded in RNA (mRNA) is decoded into a polypeptide is one of the exciting discoveries of biology. It is often referred to as deciphering the genetic code. With only four biochemical letters (A,G,C,U) a one-letter code could not unambiguously encode 20 amino acids. A two-letter code could encode only 4 X 4 = 16 amino acids and is still not enough. So, a triplet code, based on three biochemical letters or nucleotide bases could make up to 4 X 4 X 4 = 64 codons. This is the minimal that will be required to code for 20 or so different amino acids.

The discovery of the genetic code became possible through the significant contributions of Francis H.C. Crick, Severo Ochoa, Marshall W. Nirenberg, Har Gobind Khorana and J.H. Matthei in early 1960s. For this work, Har Gobind Khorana shared Nobel Prize in 1968 with Nirenberg and Holley. They used synthetic RNA homopolymers such as poly U, poly-C, poly A, and poly G or copolymers such as poly UG, AC, etc. This RNA was used in a test tube protein synthesis system with one of 20 amino acids in labelled form. Thus the first code UUU coding for amino acid phenylalanine was deciphered. All the 64 possible codons can be represented through what is known as genetic code dictionary (Fig. 14.13). This clearly shows that the first and second biochemical letters remain the same for a particular amino acid, the third biochemical base can be different. Subsequent experiments established that genetic code is nonoverlapping, unambiguous, degenerate (one amino acid coded by more than one codon), and commaless (continuous).

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First Letter (5′ end)	Second Letter U	Second Letter C	Second Letter A	Second Letter G	Third Letter (3′ end)
U	UUU Phe	UCU Ser	UAU Tyr	UGU Cys	U
	UUC Phe	UCC Ser	UAC Tyr	UGC Cys	C
	UUA Leu	UCA Ser	UAA STOP	UGA STOP	A
	UUG Leu	UCG Ser	UAG STOP	UGG Trp	G
C	CUU Leu	CCU Pro	CAU His	CGU Arg	U
	CUC Leu	CCC Pro	CAC His	CGC Arg	C
	CUA Leu	CCA Pro	CAA Gln	CGA Arg	A
	CUG Leu	CCG Pro	CAG Gln	CGG Arg	G
A	AUU Ile	ACU Thr	AAU Asn	AGU Ser	U
	AUC Ile	ACC Thr	AAC Asn	AGC Ser	C
	AUA Ile	ACA Thr	AAA Lys	AGA Arg	A
	AUG Met START	ACG Thr	AAG Lys	AGG Arg	G
G	GUU Val	GCU Ala	GAU Asp	GGU Gly	U
	GUC Val	GCC Ala	GAC Asp	GGC Gly	C
	GUA Val	GCA Ala	GAA Glu	GGA Gly	A
	GUG Val	GCG Ala	GAG Glu	GGG Gly	G

Fig 14.13 *A code dictionary depicting all the 64 possible codons. Note the initiation and 3 termination codons*

14.8 MOLECULAR MECHANISM OF MUTATION

With the identification of biochemical nature of gene as a defined segment of deoxyribose nucleic acid or DNA, mutation mechanisms were worked out at molecular level. We know that in DNA,specific sequence of bases forms the reading frame in the form of genetic code for a gene. Mutations are brought about by two main mechanisms that alter the reading frame:

(i) Substitution and

(ii) frame-shift mutations.

In substitution, one base is replaced by another base. Addition or deletion of few bases changes the reading frame and results in frame-shift mutations (Fig. 14.14). For example, the

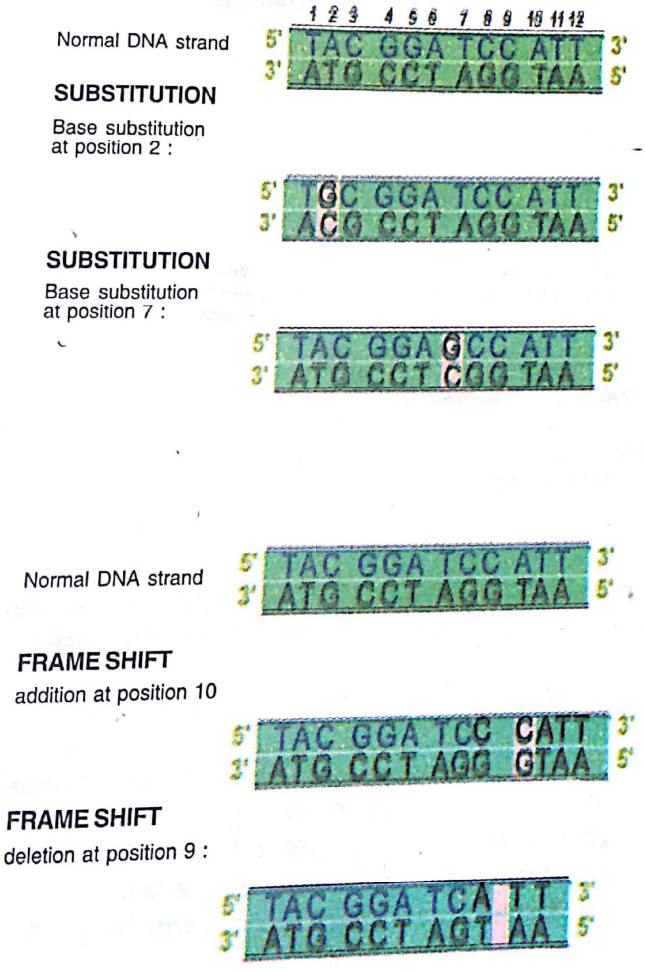

Fig 14.14 Molecular mechanisms of gene mutation

addition of T in the sentence THE FAT CAT ATE THE RAT may read as THE TFA TCA TAT ETH ERA T changing the meaning altogether. Similarly, the deletion of letter F from the same sentence shifts the reading frame of that message, producing the message THE ATC ATA TET HER AT which is meaningless. A specific base sequence within the gene is crucial for it to express a particular phenotype. Any change in the base sequence may change the codon and thus leads to altered expression or mutation. Base substitutions can also create mismatches. Each living cell possesses repair mechanisms to rectify such mistakes. Only when these mechanisms are defective, then mutations are effected.

14.9 REGULATION OF GENE EXPRESSION

In the last section, you have seen as to how the genetic information stored in DNA or gene is expressed through the process of protein synthesis. However, if we analyse the concentration of different proteins, even in a simple bacterial cell like *E.coli*, vast differences can be seen. While some proteins may be synthesised in as few as 5 to 10 molecules, others may be produced in as many as 100,000 copies per cell. This suggests that gene expression is regulated. It was known since 1900 that lactose-metabolising enzymes are synthesised by yeast only when it is grown in the presence of this sugar. Soon, bacteria were shown to adapt to their chemical environment by synthesising certain enzymes depending upon the substrate present. Such enzymes are called **inducible enzymes**. In contrast, some enzyme types are synthesised all the times irrespective of the chemical constitution of the environment; these are described as **constitutive**. Another type of regulation is revealed when the end product of a biosynthetic pathway such as an amino acid is provided in the medium. Under such a condition, the internal biosynthesis of the said amino acid is stopped. This regulation is referred to as **repressible**. Regulation can also be described as negative or positive control. Under negative control, the product of the regulatory gene shuts off the expression of the said gene(s). Under positive control, on the other hand, the product of the regulatory gene activates transcription of the concerned gene(s). The regulatory mechanisms play a very important role in the life of a cell as they prevent any unwanted gene expression thus ensuring efficient energy management.

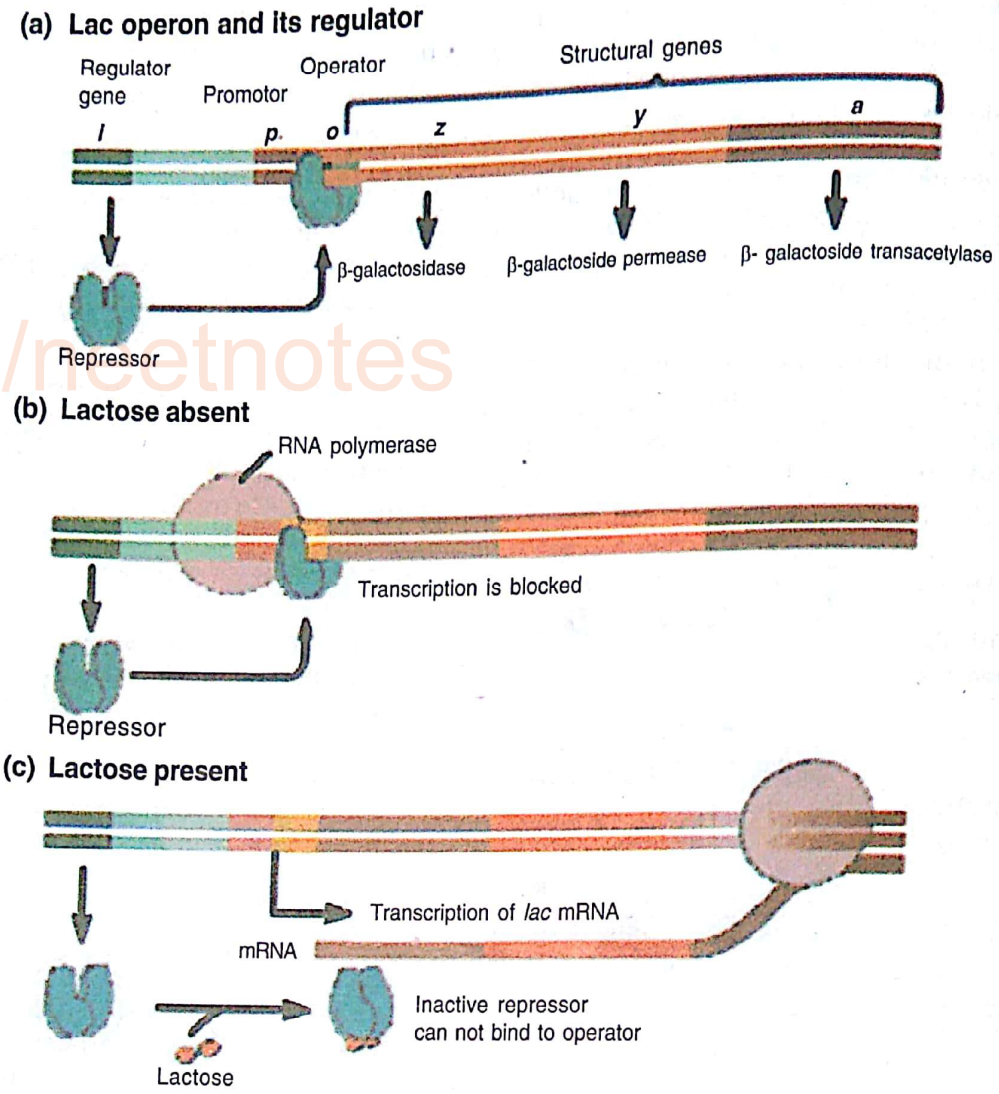

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Fig 14.15 *Inducible control regulation of lac operon in E.coli. (a) the genetic organisation of lac operon and its regulatory elements, (b) the situation in the absence of lactose, (c) the situation in the presence of lactose*
Note that the genes are expressed only when required, i.e., when lactose is present

Inducible Control

The classical example of regulation of gene activity was first proposed by Francois Jacob and Jacques Monod in 1961 while studying lactose metabolism in *E. coli*. By analysing a number of mutants, all defective in lactose metabolism, they came to the conclusion that a group of genes are expressed and regulated together as a unit that they called **operon**. In this case, the operon consists of three structural genes *Z, Y,* and *A*, as well as the adjacent sequence referred to as operator region (fig 14.15) . Subsequent to their original proposal, another region ahead of operator was identified as the promoter. You have read about the promoter in the earlier section. A regulator gene *lac i* also exists which controls the transcription from the operon by producing a repressor molecule.

The regulatory mechanism works on the basis of whether lactose is absent or present. When cells are grown in absence of lactose, the repressor molecule produced by *lacI* binds with the operator region switching off the operon. This is logical because when lactose is not present,

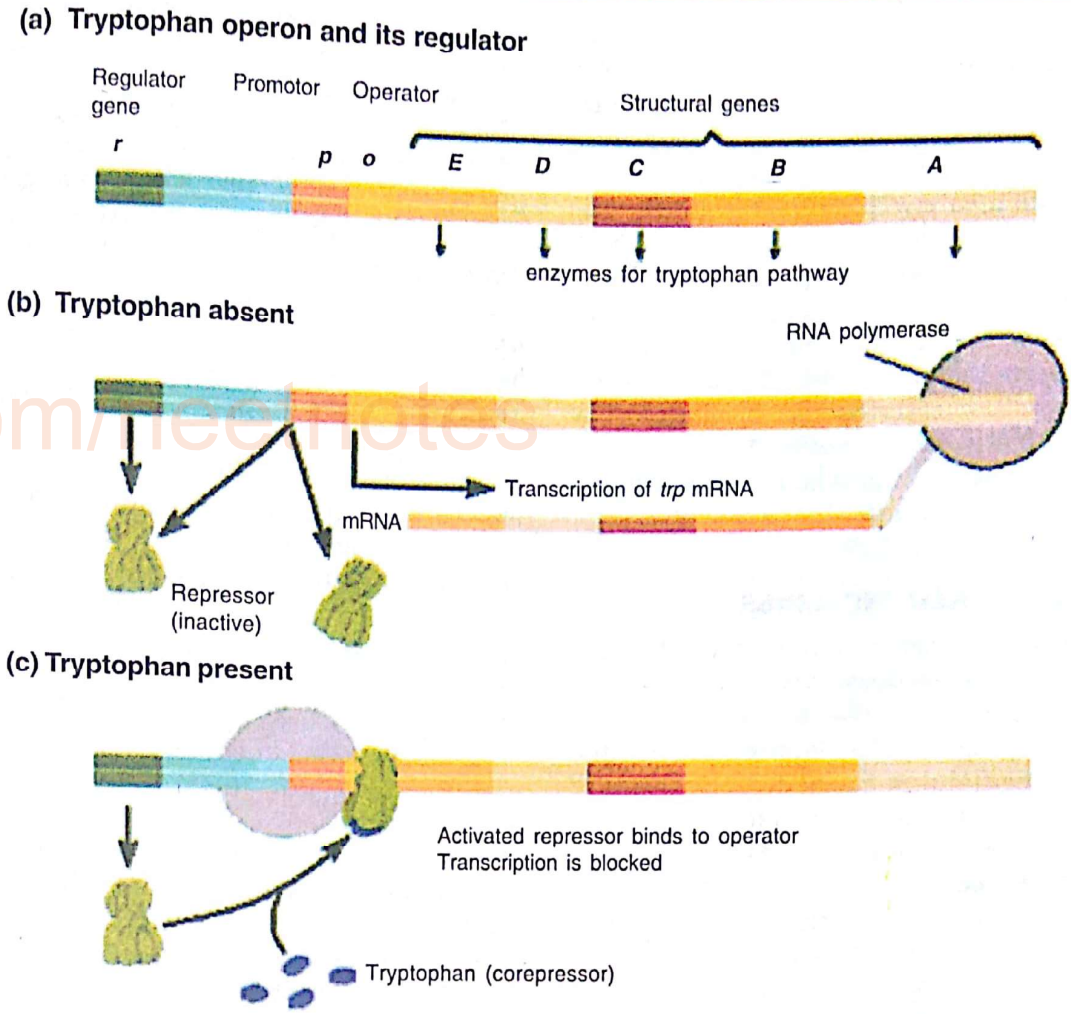

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Fig 14.16 *Repressible control of tryptophan biosynthetic genes in E.coli. (a) Genetic organization of trp operon and its regulatory gene, (b) Expression of trp genes in absence of external tryptophan, (c) Expression of trp genes in the presence of tryptophan*
Note that like in lac operon, trp genes are turned on only when required

the enzymes which process the lactose and are coded by three structural genes are also not required. However, when lactose is present, it acts as inducer. The inducer binds to repressor and modifies its structure in such a way that repressor cannot bind to operator. The three structural genes could now be transcribed and translated (Fig. 14.15). The RNA produced from the operon is polycistronic mRNA. The *lac* operon is the best-understood regulatory mechanism both in genetic and biochemical terms.

Repressible Control

If we compare the regulation of genes involved in biosynthesis of amino acids and other essential macromolecules, the situation is very different. Jacob and Monod had shown that when tryptophan is present in the growth medium, the wild-type *E. coli* cells repress the synthesis of enzymes of the tryptophan biosynthesis pathway. Enzyme repression again reflects upon the economy exerted by the cell.

Further investigations by them revealed that five genes situated close to each other on *E. coli* chromosome are involved in tryptophan biosynthesis. They proposed that these five, *trpE, D, C, B* and *A* are the structural genes, and like *lac* operon, preceded by a regulatory region constitute *trp* operon (Fig.14.16). The regulatory region here is divided into promoter, operator and a leader sequence in which the

latter is transcribed but not translated as part of the *trp* structural gene products. The operon is regulated by a distantly placed regulatory gene *trpR*. In the absence of external tryptophan, the repressor produced by *trpR* is inactive, which cannot bind to *trpO* (operator), thus allowing the transcription of polycistronic mRNA. In the presence of tryptophan, however, tryptophan binds to repressor converting this complex into active repressor that binds to operator, stopping all transcriptional activities. Such a repression of the operon, once again can be classified as negative control. The *trp* operon and in fact other amino acid biosynthesis operons are subjected to other types of control as well.

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14.10 HOUSE-KEEPING GENES

From the ongoing discussion it should become clear that gene expression is extensively regulated based on the environment. In multicellular highly differentiated organisms, there are regulations exerted even at tissue level. For example, some genes may express in liver, some in kidney, and still others in reproductive tissues. In contrast to such genes, there are common sets of expressed gene functions that are needed in all cell types. Such genes are known as house-keeping genes and their function is referred to as house-keeping or constitutive activity.

14.11 DIFFERENTIATION AND DEVELOPMENT

While talking of bacteria we usually refer to a single-celled organism with little or no differentiation. All higher plants and animals, on the other hand, are constructed from a large variety of cell types. You must be aware that such multicellular organisms also originate from a single cell, the zygote that forms as a result of fertilization. Different cell types must arise from this zygote through a highly ordered and coordinated way, a process known as differentiation. The instructions for how a fertilized egg will develop into an adult are all written in the linear sequences of the bases in the DNA or gene. This genetic information is expressed in a regulated manner not only in space and time but also allows cell-to-cell interactions to produce different body parts.

The role of nucleus in differentiation can be easily understood by nuclear transplantation experiments such as those carried out in the creation of a clonal frog. In these experiments, diploid nuclei from differentiated cells were transplanted into unfertilized eggs whose haploid nuclei were inactivated before hand. When such genetically diploid eggs were artificially induced to divide and grow, occasionally they formed adult frog whose chromosomal constitution was exactly like the donor. Although nuclei derived from larval intestinal cells could support clonal reproduction, those of adult frog do not. This suggests that pluripotency (ability to dedifferentiate completely) may be lost upon maturation. Similarly, if nerve cells are grown outside their normal cellular environment, they produce similar cells. With higher plants, however, the situation is often opposite. A complete plant can be regenerated even from the differentiated cells of the mature plant (totipotency).

As in bacteria, in multicellular organisms, all genes do not function at all times. Mechanisms must exist therefore, to control which genes will act and when, and these controls are much more complex than what we have seen in bacteria.

A wealth of information has been collected by using the fruit fly, *Drosophila*, as a working model and the developmental mutants that have been isolated. It is very clear that during development, a single fertilized egg gives rise to cells that have different developmental fates. This asymmetry may be a part of the egg itself in terms of distribution of its cytoplasmic components as in *Drosophila* or may be initiated in the initial division cycles (as in mammals). It is logical to suppose that each cell type is characterised by its pattern of gene expression or by the particular gene products that it produces. In *Drosophila*, many types of genes have been identified by mutation and analysing their functions.

These genes not only regulate one another but perhaps the target genes also that code for structural proteins. Many of these genes possess conserved motifs, the commonest of which are known as **homeobox**. Homeoboxes are distributed in other eukaryotes also, such as worms, frogs and mammals, suggesting that they may be characteristics of all animals.

14.12 CANCER AND ONCOGENES

You have studied in the earlier Unit that each somatic cell undergoes a precisely programmed cell cycle. This, in fact, controls the division and growth of a cell. This is reflected in a defined life span of not only the individual somatic cells but also the organism. One notable exception is provided by cancer cells (Fig.14.17). Cancer is a disease of cells, in which the controls that normally restrict cell proliferation do not operate.

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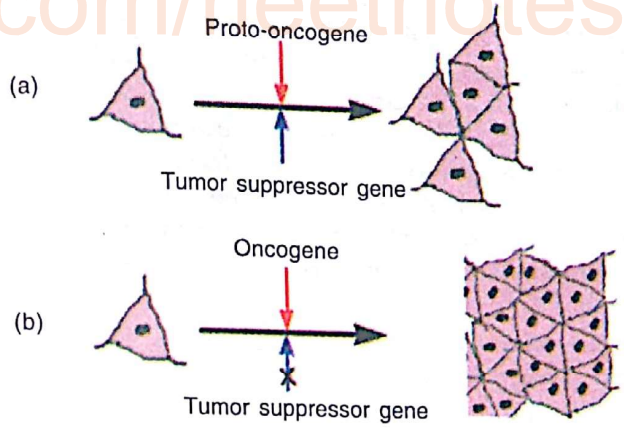

Fig 14.17 *Regulation of normal cell divisions*
(a) Cell division regulation by proto-oncogenes and constraints imposed by tumor suppressor genes
(b) Transformation of a normal cell into cancer cell if this regulation is upset

There are three major differences between the cancer cells and the normal cells.

(i) Unlike the normal cells, cancer cells are *immortalized* and show indefinite growth.

(ii) Since they are derived from normal cells, they seem to arise through *transformation* in which normal constraints of growth are bypassed.

(iii) At some stage after transformation they acquire the ability to *metastasize*, that is, they become capable of moving and invading normal tissue and find a new place away from the original site to form cancers.

When cells derived from a vertebrate are grown in culture they require a solid support to attach and essential growth factors or serum. Their growth is inhibited by cell to cell contact and they show characteristic cytoskeletal organisation. Thus, in a culture these cells grow as a monolayer. The tumor cells, show changes in some or all of these properties. Besides these, multiple genetic changes occur during the transformation of a normal cell to cancer cell, a process termed as **oncogenesis**.

Such a transformation could be caused spontaneously or by the agents known as carcinogens that promote them. The carcinogens include agents such as radiations, some chemical compounds as well as certain viruses. Currently two types of genes have been identified, whose mutations result in cancerous transformations.

(i) **Oncogenes** : Oncogenes act to stimulate cell division. Interestingly, these genes have normal cellular counterparts, called proto-oncogenes (**c-onc**) which are involved in normal cell functions. Mutations of proto-oncogenes lead to oncogenes. Many tumor causing viruses may carry similar onco gene (**v-onc**) sufficient to induce carcinogensis.

(ii) **Tumor Suppressor Genes** : As the name suggests they normally suppress the tumors and the disease appears only when the appropriate active gene is lacking or both the alleles are lost. By function, they usually impose some constraints on cell cycle. Their absence leads to the release of these constraints, leading to induction of tumors.

SUMMARY

Experimental work on biochemical and molecular nature of hereditary material conclusively demonstrated that DNA (and not protein) is indeed the genetic material. Using information gathered by many scientists, Watson and Crick worked out the biological implications and suggested a double helix model of DNA. In this, two DNA strands are wound together around each other and run in antiparallel fashion. The backbone of the molecule consists of alternating deoxyribose sugar and phosphate, and the nitrogenous bases are stacked inside. The two strands are held together by hydrogen bonding between adenine (A) and thymine (T), and guanine (G) and cytosine (C). This model also suggested the way DNA can be replicated, a prime requirement for genetic material. Replication takes place semi-conservatively, in which the two strands act as templates to allow the synthesis of new strands, which have a complementary base sequence. Replication is achieved with the aid of several enzymes, and starts at a specific point, known as origin of replication. Since the main DNA replicating enzyme acts only in 5' → 3' direction, one strand is continuously replicated but in the other, DNA is synthesised in small fragments called Okazaki fragments. The process is known as semi-discontinuous. The DNA polymerisation requires a small RNA primer also synthesised by a special enzyme, primase. Primers are subsequently removed and the gaps are filled by DNA polymerase I. Okazaki fragments are later joined by the enzyme DNA ligase.

The idea that the genes control the metabolism was available by the early part of the last century, and the same was confirmed by mid 1940s through the work of Beadle and Tatum. This and several other information established that the control is brought about by directing the synthesis of proteins which have a variety of functions. The protein sysnthesis consists of two steps, transcription and translation, which constitute the fundamental processes essential to the expression of genetic material. Transcription leads to the synthesis of three species of single-standard RNA under the guidance of the enzyme RNA polymerase from a DNA template. The three types of RNA have different functions in protein synthesis. The mRNA carries the message from the gene or the DNA, rRNA along with some proteins constitutes the ribosomes, the site at which translation takes place and tRNA transfers the required amino acid to the ribosome during translation. Both, transcription and translation can be subdivided into initation, elongation, and termination, and like replication rely on complementary base pairing. The two processes are more complex in eukaryotes where primary mRNA transcript is modified in various ways.

The genetic information is stored in the DNA in the form of a code, which is triplet, degenerate, unambiguous, non-overlapping and commaless in nature. The complete code dictionary consists of 64 possible codes, of which 61 codons code for 20 essential amino acids including the initiation codon, and 3 are meant to terminate translation. Any mutational change may result into a new codon coding for different amino acids. The expression of genes is highly regulated both by the external and the internal environments. This checks any wasteful synthesis of proteins and enables the cell to conserve energy. The synthesis of inducible class is initiated only when their substrate is available, whereas the synthesis of repressible class is stopped if their products are supplied. Also the regulation can be classified as negative or positive control. Besides repression, many biosynthetic enzymes are regulated by additional control. There are enzymes which are required by the cell althrough and are called constitutive.

The regulation of gene expression in eukaryotes is more complex, which obviously is a reflection of their multicellularity, larger genome size and other aspects of gene expression. The process of development and differentiation and expression of specific sets of gene such as oncogene have served as models for gene regulation in higher organisms. The whole

process of development and differentiation is based on differential gene expression, as the basic genetic information carried by each somatic cell in a multicellular organism is same. Though tissue specific gene expression is known, there are some common sets of expressed genes also. Such genes are known as house-keeping genes.

An aberration in the normal plan of cellular growth and its regulation can lead to carcinogenesis or induction of cancer. In this process, the normal cells are transformed to a state where they lose normal constraints of growth. A cell possesses proto-oncogene or *c-onc* in which alteration can lead to their transformation into cancerous cells. Many tumor causing viruses may carry similar c-*onc* gene (*v-onc*) which may be sufficient for carcinogenesis. Similarly, a normal cell may carry tumor suppressor genes and the loss of the same may result in tumor induction.

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EXERCISES

1. Which of the nucleotide compositions will be possible, if DNA is double stranded?
   (a) All A; (b) Only A and T; (c) Only C and T; (d) Only A and G; (e) only A,G and T.
2. Given below is the transcribed strand of the DNA duplex:
   3'– TAC CGA TCC GAG CTG –5'
   (a) Draw the complementary DNA polynucleotide chain
   (b) Construct the RNA molecule, which will be transcribed.
3. From the following DNA sequence representing a part of the gene, derive:
   (a) the RNA transcript;
   (b) the processed mRNA (assuming that all the codons containing a C represent the intron DNA);
   (c) the number of amino acids it can code for:
   TACCCCCAC GAGTTATATATACGG GGGCATCATATG
4. Consider DNA molecules which contain most of its nitrogen in the form of ^{15}N (heavy isotope). Such molecules are allowed to replicate in an environment, where the nitrogen source is ^{14}N. What will be the proportion of the DNA molecules that will contain some ^{15}N after: (a) one round of replication; and (b) two rounds of replication?
   Provide experimental evidence for the semi-conservative mode of replication of DNA.
5. Given below is sequence of the processed mRNA ready for translation:
   5'– AUG CUA UAC CUC CUU UAU CUG UGA–3'
   (a) How many amino acid residues will make up the polypeptide corresponding to this mRNA?
   (b) How many different tRNA molecules would be necessary to translate this mRNA?
6. What was the rationale of using ^{32}P and ^{35}S by Hershey and Chase? Instead, if we use radiolabelled C and N, will the results be any different?
7. List three main differences between DNA and RNA?
8. Which molecule bears codons and which molecule anticodons?
9. What are the three essential requirements of the genetic materials?
10. How did Hershey and Chase prove that DNA is the genetic material?

Chapter 15

Genetic Engineering, Cloning And Genomics

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In Chapter 14, you have learnt about DNA replication, transcription and translation, and how gene expression is regulated. In this chapter, you will learn about the tools and technologies of molecular biology for cleaving and rejoining DNA sequences from two or more different organisms. These genetically modified DNA fragments are termed as **recombinant DNA** molecules. These can be cloned and amplified virtually to an unlimited extent. Application of DNA manipulation biotechnology ranges from cloning genes to cloning organisms including transgenic microbes, agriculturally important crops and farm animals. Using these, we can get genetically modified food products, human gene products, pharmacologically and therapeutically usedful products. Gene replacement or gene augmentation therapy for hereditary diseases can provide treatment to some of the presently otherwise incurable disorders. DNA fingerprinting, a reliable technique, has immense application in forensic investigations. And finally, we now have human genome deciphered revealing the blue print of our life itself !

15.1 GENETIC ENGINEERING

Genetic engineering is now considered as a kind of biotechnology. It is essentially the alteration of the genetic make up of cells by deliberate and artificial means. It involves transfer or replacement of genes to create recombinant DNA. How is this done? This can be made possible because we can cut DNA molecules at specific sites to get fragments containing desirable and useful genes from one type of cell. Subsequently these genes can be inserted into a convenient carrier or vector. Now, these recombinant DNA can be put into completely different cell of a bacterium or plant or animal. This enables them to acquire useful characteristics, for example, disease resistance or to make useful and better products, such as enzymes, hormones, vaccines and so on. The techniques involved require a lot of precision and high skill.

Tools of Genetic Engineering

From isolated DNA from a cell culture with the desired gene, DNA segment can be excised by 'molecular scissors or chemical scalpels', what biotechnologists call **restriction enzymes**. Restriction enzymes, synthesised by microbes as a defense mechanism, are specific endonucleases, which can cleave double-stranded DNA. However, they can do so only at specific limited number of sites depending on the number of recognition sequences in DNA. Two examples of restriction enzymes that give either **sticky** or **blunt ends** are shown in Figure 15.1.

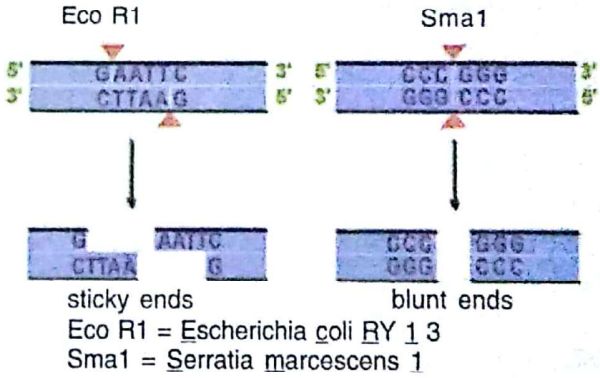

Fig. 15.1 Restriction enzymes

A **vector** is used as carrier for transferring selected DNA into cells. A plasmid with its small DNA from a bacterium is a good choice for indirect gene transfer, because it can move from one cell to another and make several copies of itself. However, artificial chromosomes from bacteria and yeast called **BACs** and **YACs**, respectively are more efficient for eukaryotic gene transfers (Fig. 15.2).

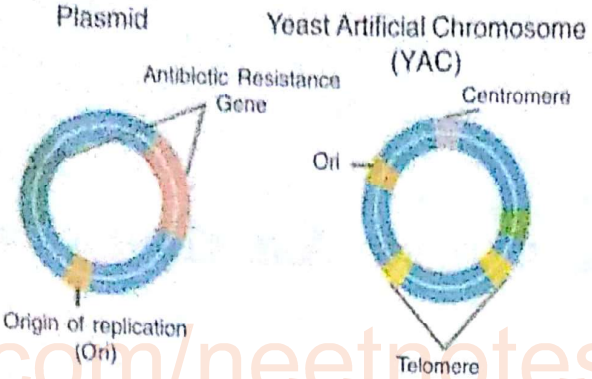

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Fig. 15.2 *Plasmid and Yeast Artificial Chromosome*

You also require DNA synthesising enzymes such as **DNA polymerase** or **Reverse transcriptase** to make DNA complementary to an existing DNA or RNA template respectivly, called cDNA (complementary DNA). Because of the universality of the genetic code, the DNA polymerases can accuratly transcribe a DNA sequence from one organisim to another organism. And finally molecular sutures, the **DNA ligase** enzymes to anneal or join DNA fragments. They do so by again forming phosphodiester bond. Any pair of fragments produced by the same restriction enzyme, from any DNA source, fruitfly to elephant, can be joined together.

Recombinant DNA Technology

Gene transfer involves essentially the following stages:

(i) Isolating a useful DNA segment from the donor organism;

(ii) Splicing (inserting) it into a suitable vector under conditions to ensure that each vector receives no more than one DNA fragment or gene to create a recombinant DNA;

(iii) Producing of multiple copies of this recombinant DNA;

(iv) Inserting this altered DNA into an appropriate recipient organism; and

(v) Screening of the transformed cells.

All that remains is to persuade the cell to begin making the appropriate gene product and then devise an economical method for its mass production. The various steps and their sequence for the production of human insulin are depicted in Fig.15.3.

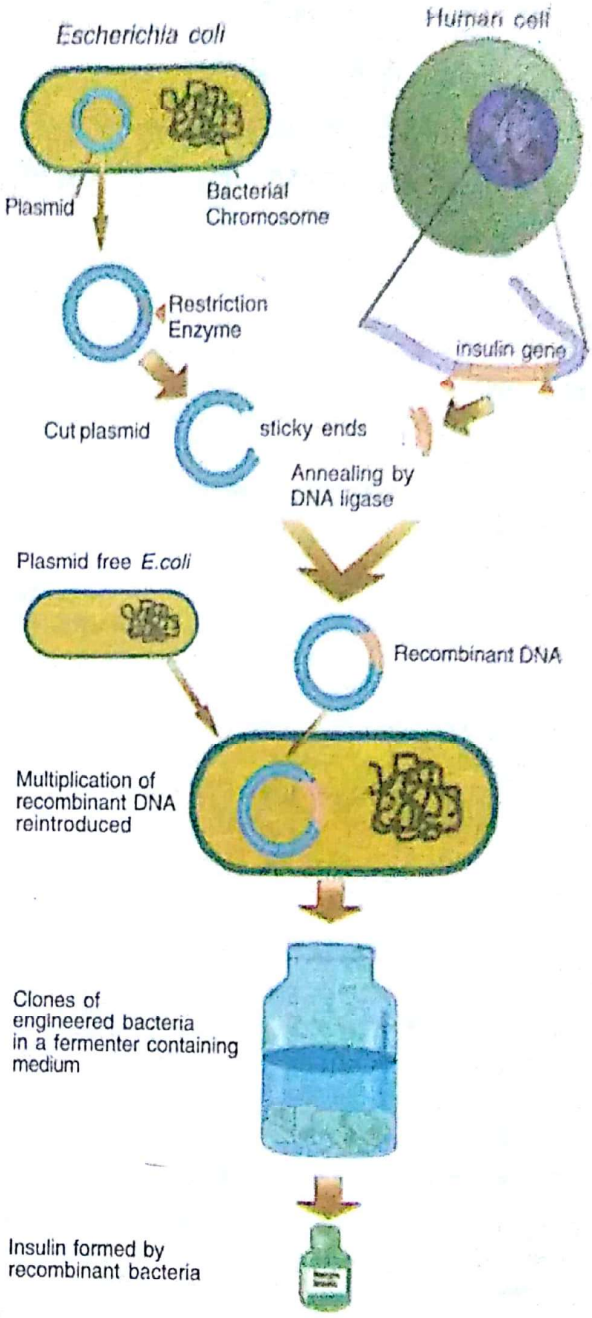

Fig. 15.3 *Steps involved in gene transfer for the production of human insulin*

Another successful example is based on the **Ti (Tumor-inducing) plasmid** which occurs in the soil bacterium *Agrobacterium tumefaciens* (Fig 15.4).

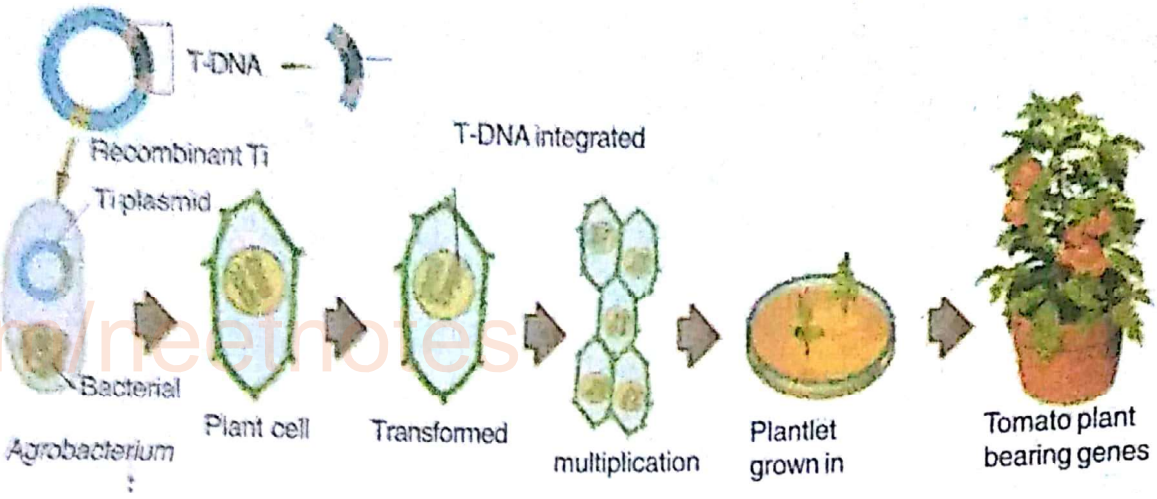

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Fig. 15.4 Agrobacterium Ti plasmid-mediated genetic transformation in plants

This bacterium infects all broad-leaved agricultural crops such as tomato, soyaben, sunflower and cotton but not cereals. It induces formation of cancerous growth called a crown gall tumor. This transformation of plant cells is due to the effect of Ti plasmid carried by the pathogenic bacterium. So for genetic engineering purposes, *Agrobacterium* strains have been developed, in which tumor-forming genes have been deleted. These transformed bacteria can still infect plant cells. The part of Ti plasmid transferred into plant cell DNA, is called the **T-DNA**. This T-DNA with desired DNA spliced into it, is inserted into the chromosomes of the host plant where it produces copies of itself, by migrating from one chromosomal position to another at random. But it no longer produces tumors. Such plant cells are then cultured, induced to multiply and differentiate to form plantlets. Transferred into soil, the plantlets grow into mature plants, carrying the foreign gene, expressed throughout the new plant.

Vectorless Direct Gene Transfer

New technologies like gene gun are also available for vectorless direct gene transfer. DNA coated onto microscopic pellets is literally shot into target cells (Fig. 15.5). Though developed for plants, this delivery technique has been used successfully to insert genes that promote tissue repair into cells near wounds, leading to a dramatic reduction of healing time.

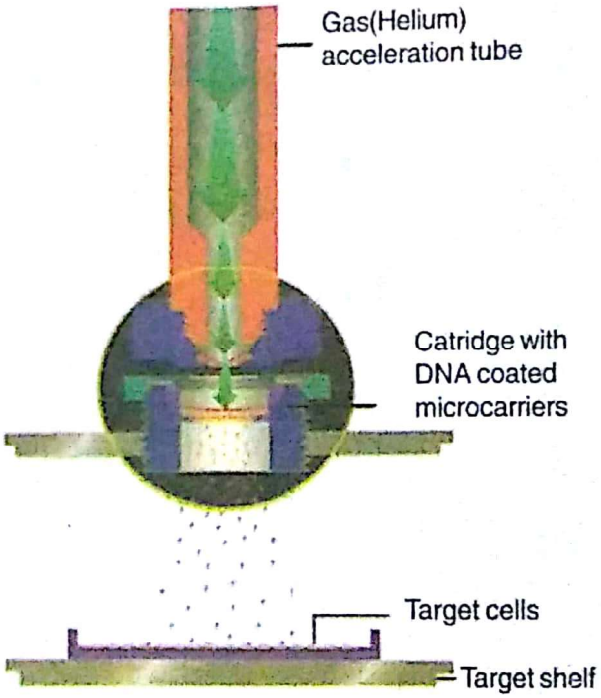

Fig. 15.5 Schematic of Gene gun

Applications of Recombinant DNA Technology

The technique of recombinant DNA can be employed in the following ways:

(i) It can be used to elucidate molecular events in the biological process such as cellular differentiation and aging. The same can be used for making gene maps with precision. You will also learn later in this chapter, how tools and techniques of recombinant DNA

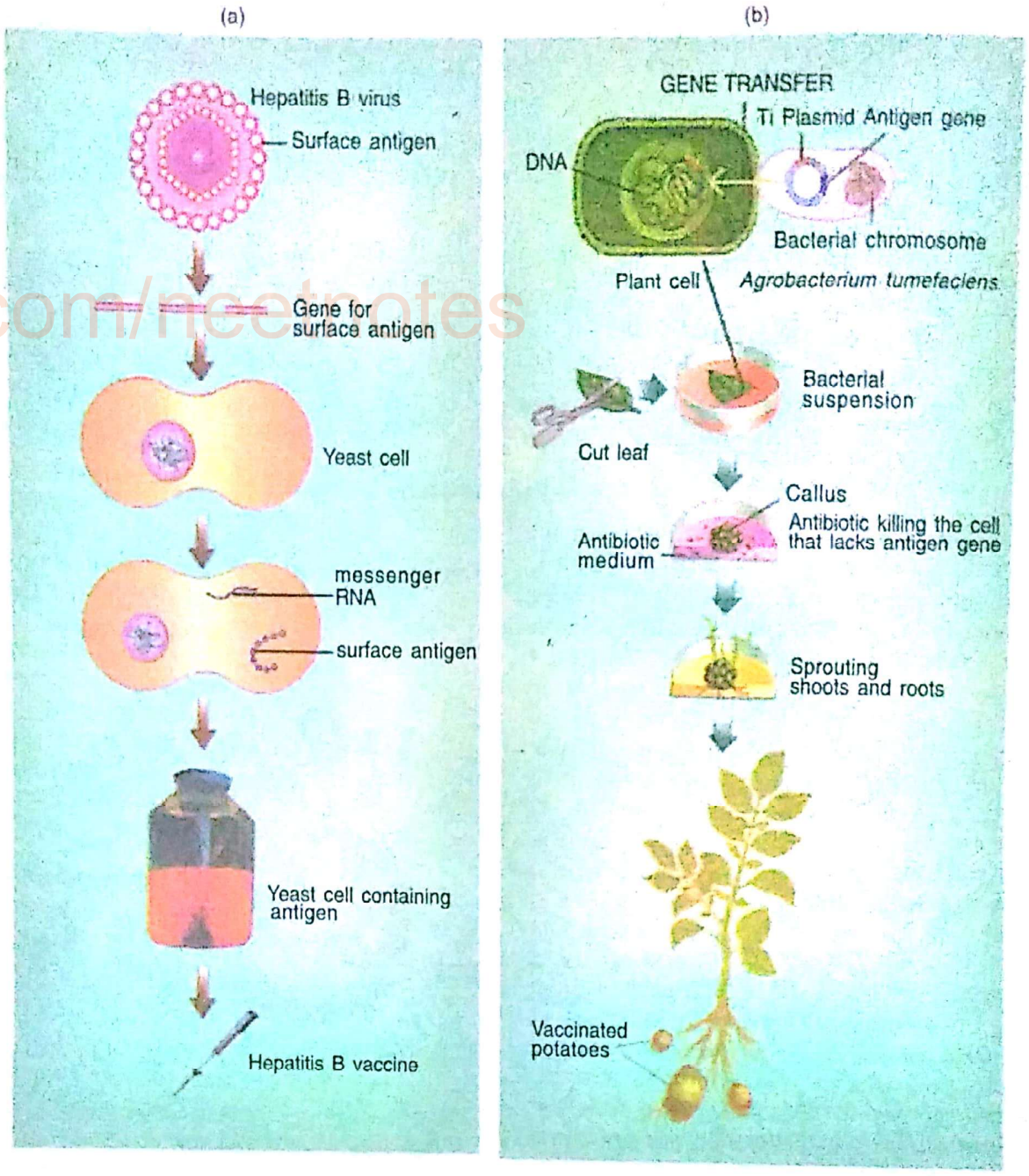

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Fig. 15.6 *Production of vaccines through recombinant DNA technology. (a) injectable hepatitis B vaccine, (b) edible vaccine*

were used to spell out the complete nucleotide sequence of genome of various organisms, including human beings.

(ii) In biochemical and pharmaceutical industry, by engineering genes, useful chemical compounds can be produced cheaply and efficiently (Table 15.1). Two examples of production of vaccine, through recombinant DNA technology are shown in Fig. 15.6. Years of testing, however, will be needed before such treatments become widely available.

Table 16.1 Applications of Recombinant DNA Products

Medically useful recombinant products	Applications
Human insulin	Treatment of insulin – dependent diabetes
Human growth hormone	Replacement of missing hormone in short stature people
Calcitonin	Treatment of rickets
Chorionic gonadotropin	Treatment of infertility
Blood clotting factor VIII/ IX	Replacement of clotting factor missing in patients with Hemophilia A/ B
Tissue Plasminogen Activator	Dissolving blood clots after heart attacks and strokes
Erythropoietin	Stimulation of the formation of erythrocytes (RBCs) for patients suffering from anemia during kidney dialysis or side effects of AIDS patients treated by drugs.
Platelet derived growth factor	Stimulation of wound healing
Interferon	Treatment of pathogenic viral infections, cancer
Interleukins	Modulation of action of immune system
Vaccines	Prevention of infectious diseases such as hepatitis B, herpes, influenza, pertussis, meningitis, etc.

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Medical Diagnosis of Diseases

Recombinant DNA technology has provided a broad range of tools to aid physicians in the diagnosis of diseases. Most of these involve the construction of probes: short segments of single-stranded DNA attached to a radioactive or fluorescent marker. Such probes are now routinely used for identification of infectious agents, for instance, food poisoning *Salmonella*, pus forming *Staphylococcus*, hepatitis virus, HIV and so on. By testing the DNA of prospective genetic disorder carrier parents, their genotype can be determined and their chances of producing an afflicted child can be predicted.

Gene Therapy

What was the potential of genetic engineering is now prospect of Gene Therapy. Gene therapy, in humans, is to replace the function of "a faulty gene" by a normal healthy functional gene. Under intensive investigation are, diseases ranging from the rare genetic diseases caused by single mutations like sickle cell anaemia to killer diseases such as Severe Combined Immuno-Deficiency (SCID). For instance, SCID is caused by a defect in the gene for the enzyme adenosine deaminase (ADA) in 25 per cent of the cases. These patients have no functioning T-lymphocytes and so they cannot mount immune responses against invading pathogens. The ideal approach for SCID treatment would be to give the patient a functioning ADA that breaks down toxic biological products. But how to accomplish such gene therapy? Lymphocytes, a kind of white blood cells are extracted from the bone marrow of a SCID sufferer. A good copy of human gene encoding this enzyme is introduced into these cells (Fig. 15.7). This is done by exploiting the properties of special viruses called retroviruses, as vector. This vector is engineered to incorporate the ADA gene that has the packing sequence but no viral genes. Co-infection of the bone marrow stem cells, removed from a SCID patient's hip bone with this vector and helper virus produces new virus particles with ADA gene inserted inside, but lacking the packing sequence. This equips these cells with a good copy of the gene, in addition to the existing faulty copy. The treated cells are then reinjected to the patient's bone marrow. Lymphocytes subsequently produced by these stem cells have a fully functional ADA gene. These cells then can play their normal role in patient's immune system. This molecular

surgical approach to introduce 'therapeutic' genes into particular tissues which can work throughout life, is confronted by many practical challenges but it offers the hope of specific therapy (Fig. 15.7).

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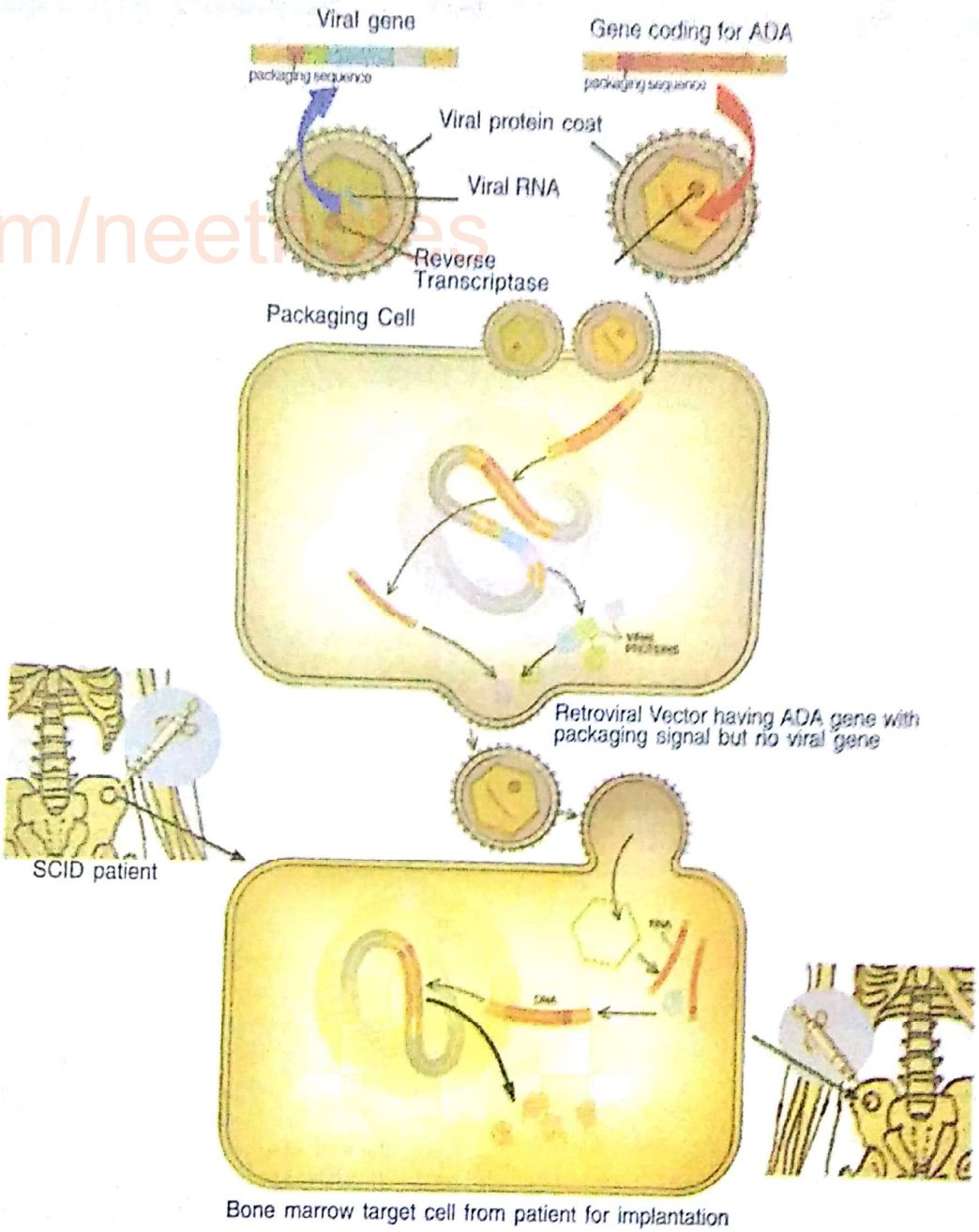

Fig. 15.7 *Gene therapy to treat SCID : To make the therapeutic vector, two retroviruses are engineered. In the first, the packing sequence on the viral RNA are removed so that this virus can not pack its RNA into a protein after it has replicated in a human cell. In the second virus, the RNA that instructs a cell to make viral protein coat is replaced with a human adenosine deaminase enzyme gene. Thus this vector is engineered to corporate ADA gene that has the packing sequance but no viral gene. A few bone marrow stem cells from deep within SCID patient's hipbone are extracted with an injection needle. These cells are put into a plastic bag. The co-infection of these cells with the altered retrovirus as well as helper virus, lacking the packaging signal, produces new virus particles with ADA gene incorporated inside. The genetically engineered bone marrow cells are then infused back to the patient's bone marrow. Some cells incorporate the gene in their DNA and start to synthesize the missing enzyme.*

15.2 CLONING

Clone means exact carbon copy or progeny of a single parent. The word "Clone" only has its meaning to living species. So two exact similar soft-drink bottles will not be clones. Clones, owing to their identical genetic make up exhibit little, if any, genetic variation. In nature, organisms like microbes and plants that reproduce asexually, produce clones. There are lots of clones in the world. Monozygotic identical twins are clones. They started off as a single cell in their mother's womb. The fertilized egg divided into two and each cell developed into a separate twin having the same genetic characteristics. "Dolly" , the most famous sheep in the world, was the first animal clone (Fig. 15.8).

Cloning is the process of producing many identical organisms or clones. Dolly was produced from a single cell from her mother. She did not have a father, because no sperm was needed. She had exactly the same genetic characteristics as her mother, the single parent.

Fig. 15.8 Dolly, the sheep developed through cloning

Microbial Cloning

Each of the genetically altered or modified microbial cell can be duplicated every time it divides. In just a few days there will be millions of cloned cells, each one carrying a copy of the original donated gene. Several improved genetically altered strains of microorganisms can be cloned in large numbers for various applications (Table 15.2).

Table 15.2 Applications of Genetically Engineered Microbes

Microbes	Applications
Escherichia coli (gut bacterium)	Production of human insulin, human growth factor interferons, interleukin and so on
Bacillus thuringiensis (soil bacterium)	Production of endotoxin (Bt toxin), highly potent, safe and biodegradable insectide for plant protection
Rhizobium meliloti (bacterium)	Nitrogen fixation by incorporating "nif" gene in cereal crops
Pseudomonas fluorescence (bacterium)	Prevention of frost damage to the plants (e.g. strawberries) on which it grows
Pseudomonas putida (bacterium)	Scavenging of oil spills by digesting hydrocarbons of crude oil
Bacterial strains capable of accumulating heavy metal	Bioremediation (cleaning of pollutants in the environment)
Trichoderma (fungus)	Production of enzyme chitinases for biocontrol of fungal diseases in plants

Cell Cloning

Cell cloning is based on the property of totipotency (in plants) or pluripotency (in animals). **Totipotency** is the potential ability of a plant cell to grow into a complete plant. In contrast, **pluripotency** is the potential ability of a cell to develop any type of the cell in the animal body, for instance, nerve cells or kidney cells or heart cells. Virtually, all plants are totipotent but in animals only fertilized egg and stem cells in the embryonic blastocyst are totipotent. Moreover in contrast to animals, there is no real distinction in plants between somatic cells and germ line cells.

Plant Cloning

Many orchids producing beautiful flowers are cloned plants. Scientists have genetically engineered agronomically important crop plants. Rapid production can be achieved using actively dividing meristematic cells. These occur at the root and shoot apices that are growth areas of the plant. You can perform this simple 'clone it yourself experiment'(Fig. 15.9)

Animal Cloning

Animal cloning is more difficult than plant cloning. This is because animal cells lose their totipotency on reaching the gastrula stage of animal development. How was then celebrity sheep "Dolly" cloned? Ian Wilmut and his colleagues at the Roslin Research Institute in Scotland, took cells from ewe's (mother sheep) udder (Fig. 15.10). As a somatic cell an udder cell is different from a skin cell or a muscle cell or a nerve cell. They managed to store these udder cells in nutrient deprived culture. This halted the starved cells from dividing, and switched off their active genes. One udder cell complete with its nucleus was selected, because this nucleus carries the mother's genetic information. Meanwhile, the unfertilized egg cell was taken from a different ewe (host mother sheep). Its nucleus was sucked out leaving an empty cell containing all the necessary machinery to produce an embryo. This cell was now ready to receive udder cell nucleus. They then fused udder cell nucleus with the empty egg cell by electrical stimulation. Now, this egg

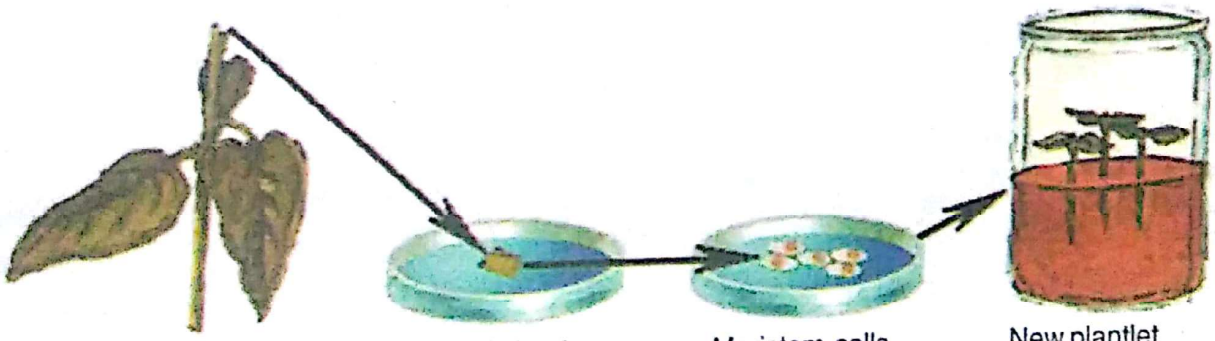

Fig. 15.9 *Cloning of a genetically engineered plant*

For the agriculture, disease-, drought-, insect pest-resistant as well as herbicide-tolerant crops have been successfully produced by gene manipulation. **Genetically Modified Food** (GMF) like vitamin A-rich rice and lysine-rich pulse seeds are now becoming components of human staple food.

cell had the mother's nucleus. When a normal or altered egg is implanted in a different female other than the egg donor, such female is termed 'surrogate mother', meaning substitute mother. The altered egg was cultured for six days. Out of many resulting embryoes, one was implanted in the uterus of the surrogate mother, where it grew

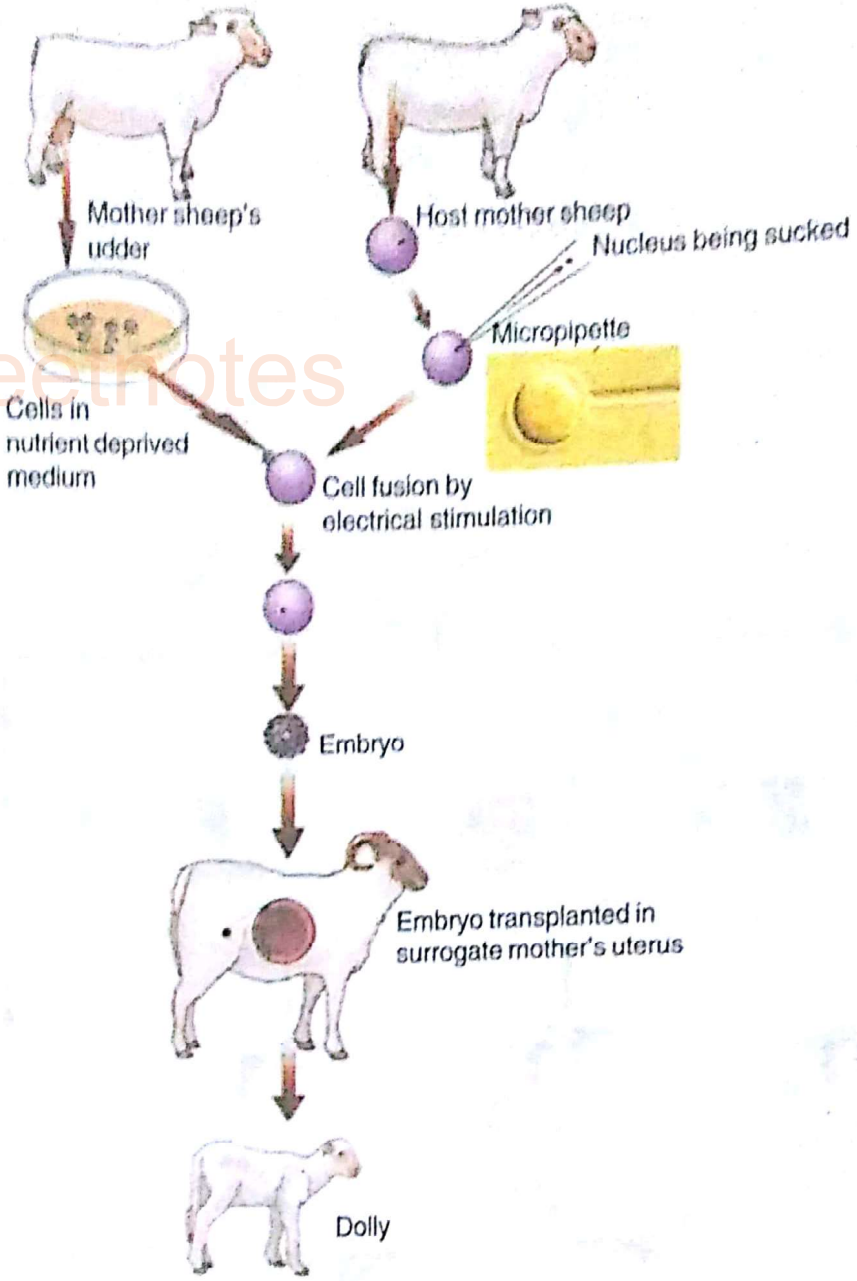

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Fig. 15.10 *Steps in animal cloning*

into a lamb. Thus, Dolly was born, genetically identical to mother sheep because her first cell nucleus came from mother's cell.

Scientists from Japan have cloned cattle in a different way. They have succeeded in growing as many as eight identical calves from one fertilized cell of their mother. How could they achieve this? The idea is straight forward, but there are several difficult steps along the way. When the mother cow has mated with the bull, she has a fertilized egg (zygote) in her womb (Fig. 15.11). This cell divides in two and then in four and then in eight. This embryo is carefully removed from the womb. The embryonic cells are then separated using an enzyme. Each isolated cell is kept in a nutrient medium, and later implanted in the womb of a different 'foster mother' cow. The foster mother's womb must accept the cell and make it grow. But each hit-and-miss step is often tricky and requires precision and skill. Each cell grows into a normal, healthy, baby calf, if every thing goes well.

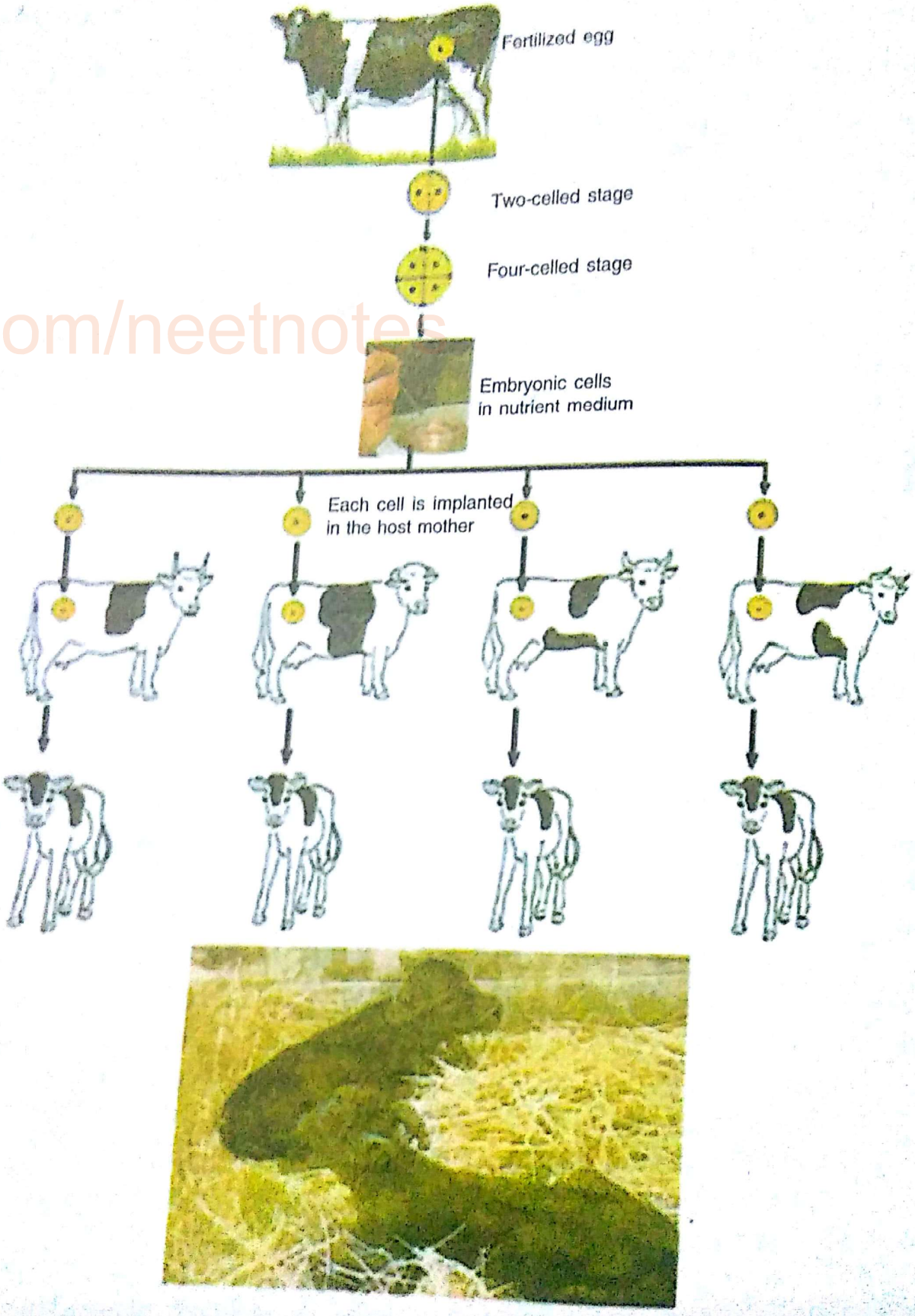

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Fig. 16.11 *Clonal calves*

15.3 TRANSGENICS

Transgenics or transgenic organisms are also called **Genetically Modified Organisms** (GMOs). They are created by incorporating into their genetic make up foreign gene(s) or extra copies of an endogenous gene or a cloned and modified gene. Transgenic microbes are made viable by cloning. You have already encountered them in Section on 'Microbial Cloning'. Transgenic crops with useful characters can be produced. For example, the gene coding for the insecticidal protein from *Bacillus thuringiensis* (Bt) has been transferred to the cotton plant. This transgenic cotton plant known as genetically modified Bt cotton is resistant to bollworm (Fig. 15.12).

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Fig. 15.12 Genetically modified Bt cotton

The transgenic GMO tomato called Flavr Savr (Fig. 15.13) has a much longer and more flavourful shelf-life than conventional tomatoes, because of delayed ripening. This is achieved by reducing the amount of pectin of cell wall degrading enzyme 'polygalacturonase' responsible for fruit softening.

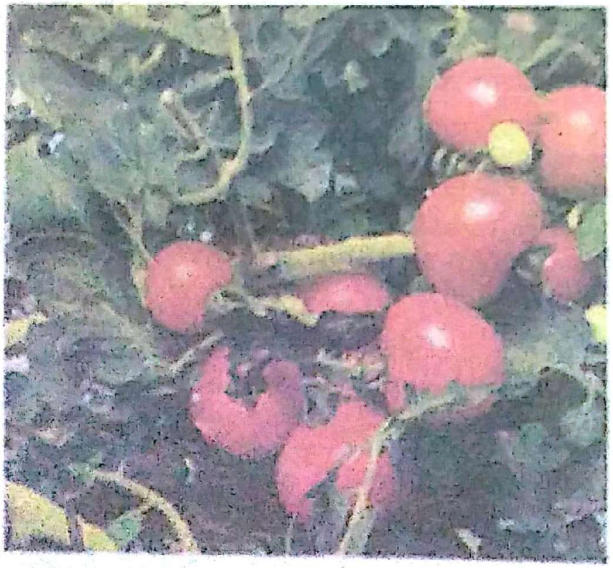

Fig. 15.13 Flavr Savr: Genetically Modified tomato

What is the difference between the two mice shown in Fig. 15.14?

Fig. 15.14 Transgenic mouse as compared to the normal mouse

One is normal in size, the other more than twice as big. The bigger is transgenic. It is larger because of expression of the gene for human growth hormone that has been introduced into it.

Transgenic cattle that have received additional gene, for instance, to increase growth rate, can be marketed more quickly. Transgenic animals have been genetically engineered with human genes in order to make products that can be used in human medicine. Here are some examples: Transgenic cattle (cow, sheep and goat) have been produced with the intention of increased milk yield, as well as, therapeutic human proteins. The cattle secrete these useful proteins in their milk from where it can be 'harvested'. Transgenic animals can be given human genes so that their organs carry human antigens.Attempts have been made to use transgenic pig organs like heart, kidney, pancreas for transplanting into humans. Surprisingly, these are not rejected (Table 15.3).

Table 15.3 Transgenics and their Potential Applications

Transgenic	Useful applications
Bt Cotton	Pest resistance, herbicide tolerance, and high yield
Flavr Savr Tomato	Increased shelf-life (delayed ripening) and better nutrient quality
Golden Rice	Vitamin A-rich
Cattles (cow, sheep, goat)	Therapeutic human proteins in their milk
Pig	Organ transplantation without risk of rejection

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15.4. DNA FINGERPRINTING

Every individual organism is unique. Like the fingerprints, each person has a unique DNA fingerprint. Unlike a conventional fingerprint that occurs only on the fingertips and can be altered by surgery, a DNA fingerprint is the same for every cell, tissue and organ of a person. It cannot be altered by any known treatment. The ideal way to distinguish an individual from other people on earth would be to describe his or her entire genomic DNA sequence as discussed later. This idea is clearly not practical. So, we look for genes that are highly polymorphic, that is, DNA sequences that have multiple alleles in the human population, and therefore different in different individuals.

The technique of DNA fingerprinting was pioneered and perfected by British genetist Dr. Alec Jeffreys. In 1984, he had developed this process to visually identify DNA found between the genes in the hope that it would reveal markers for inherited diseases and lead to early treatment. He had no idea at the time that his new technique would help to solve a murder case by comparing DNA's.

Principle of DNA Fingerprinting

Important for DNA typing or profiling or fingerprinting are short nucleotide repeats that vary in number from person to person, but are inherited. These are the **Variable Number Tandem Repeats or VNTRs** (Fig. 15.15). The VNTRs of two persons may be of the same length and sequence at certain sites, but vary at others. In this example, a child might inherit a chromosome with six tandem repeats from the mother and the same tandem repeated four times in the homologous chromosome inherited from the father. Note that the half of VNTR alleles of the child resemble that of the mother and half that of the father.

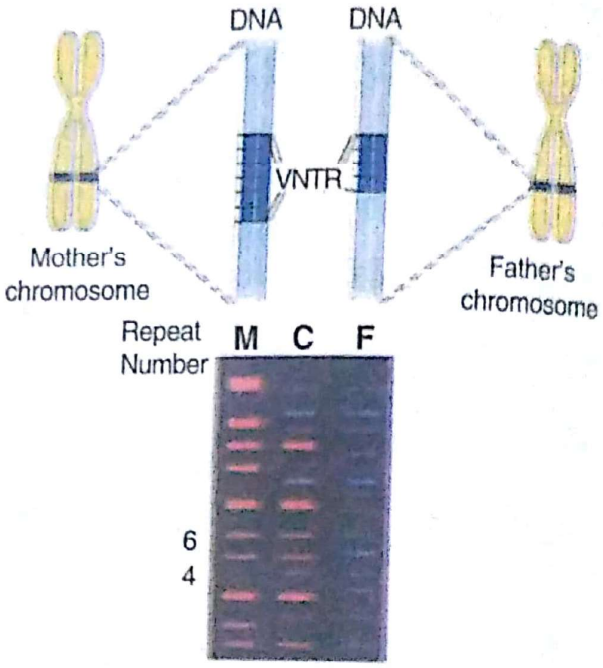
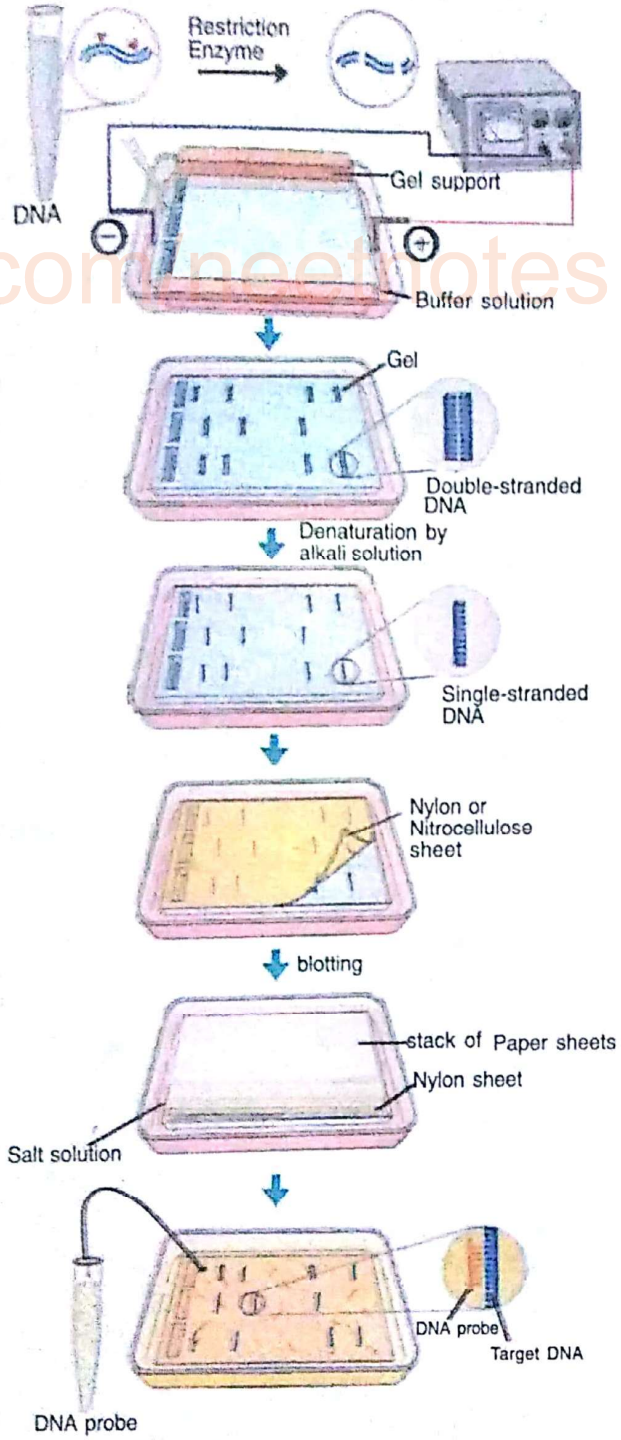


Fig. 15.15 *Variable Number Tandem Repeats (M = mother, F = father, C = child)*

Technique for DNA Fingerprinting

For DNA fingerprinting, only a small amount of tissue like blood or semen or skin cells or even follicle root of the hair is needed. Typically DNA content of about 100,000 cells or about 1 microgram is sufficient. The procedure of DNA fingerprinting involves the following major steps:

(a) DNA is extracted from the cells.
(b) If the content of DNA is limited, DNA can be amplified by making many copies of it using PCR or Polymerase Chain Reaction.
(c) DNA is cut into fragments with site recognising restriction enzymes (Fig. 15.16) for **restriction fragment length analysis.**



(d) Chopped DNA fragments are introduced and passed through electrophoresis set up containing agarose polymer gel. The separated fragments can be visualised by staining them with a dye that fluoresces under ultraviolet radiation.

(e) Double-stranded DNA is then split into single-stranded DNA using alkaline chemicals.

(f) These separated DNA sequences are transferred to a nylon or nitrocellulose sheet placed over the gel. This is called 'Southern Blotting' after its inventor E.M. Southern.

(g) The nylon sheet is then immersed in a bath, and probes or markers that are radioactive synthetic DNA segments of known sequences are added. The probes target a specific nucleotide sequence which is complementary to VNTR sequences and hybridizes them.

(h) Finally, X-ray film is exposed to the nylon sheet containing radioactive probes. Dark bands develop at the probe sites which resemble the bar codes used by scanners to identify items.

Applications of DNA fingerprinting

This technique is now used to :

(i) identify both victims and killers in forensic laboratories,

(ii) determine paternity, that is, who the true biological father or mother is of a child,

(iii) verify whether a hopeful immigrant is, as he or she claims, really a close relative of already an established resident, and

(iv) identify racial groups to rewrite biological evolution.

Fig. 15.16 *Technique of DNA fingerprinting*

15.5 GENOMICS

All the DNA in the cells of an organism, say for our cells from skin or muscle or brain and everything including genes, is its **genome**. Our body contains 100 million cells of over 260 different kinds. However, in most cases complete set of instructions in each is the same, needed to make a whole new human being. In all, there are 23 different chromosomes containing packed DNA in a haploid set of human genome. Additional DNA is in cell's mitochondria which is inherited from one's mother.

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Human Genome Project

An ambitious international Human Genome Project began in 1990. The goals of this project are:

(i) To develop ways of mapping the human genome at increasing fine level of precision,

(ii) To store this information in databases and develop tools for data analysis, and

(iii) To address the ethical, legal and social issues that may arise from this project.

Fig. 15.17 *The Human Genome*

Human Genome

Finally we have our genome deciphered in the form of genetic script of human life (Fig. 15.17). According to estimates, there are about three billion of biochemical letters: nucleotide base pairs in human genome.

Revelations of Genome

The details of the findings of our genome have revealed some startling facts. Being the most complex organism, we were expected to have more than 100,000 genes. Instead, the human gene count is much lower than expected, approximately 30,000 to 40,000 genes (Fig. 15.18).

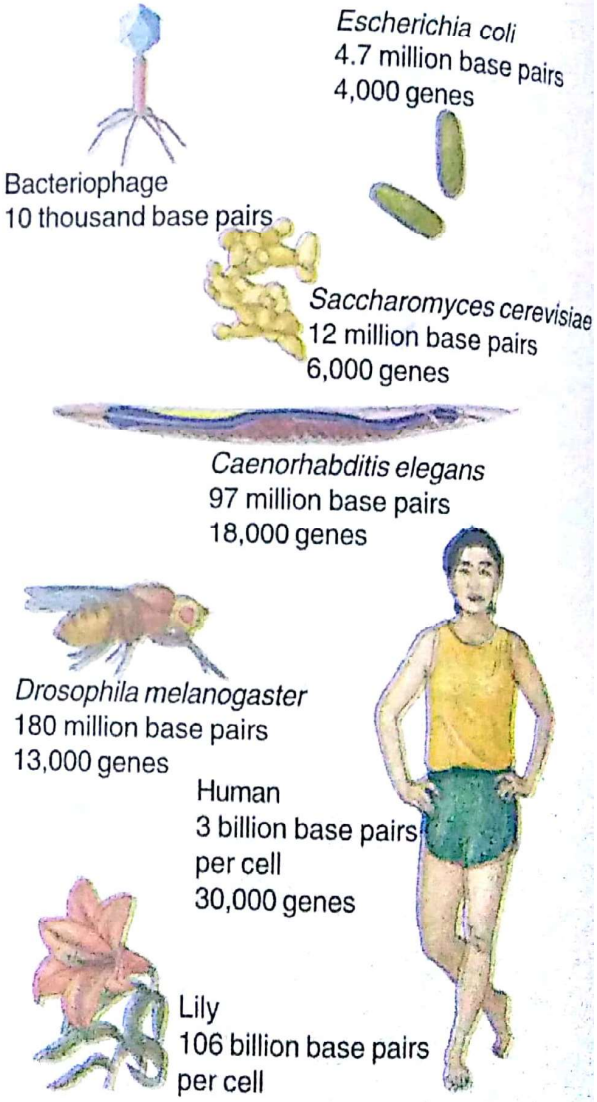

Fig. 15.18 *Comparative genomes*

This is around the same in mice. Nine-tenths of our genes are identical to that of the mouse, a favoured experimental animal. We have more than twice as many genes as fruit fly, *Drosophila melanogaster*, and we have only six times more genes than bacterium, *Escherichia coli*.

Another surprise is that we are incredibly 99.9 per cent identical with each other at the DNA level. Genomically speaking, every alive human being is exactly the same, and even bacteria are our cousins in code. Further most of our genetic differences are shared among all ethnicities and races spread into the geographic regions around the world. Different human genes vary widely in the length often over thousands of base pairs. While ß-globin and insulin gene are less than 10 kilo base pairs, the gene responsible for Duchenne Muscular Dystrophy on 'X' chromosome is made up of 2400 kilo base pairs. This is probably the longest gene known.

But why does a lily which produces beautiful flowers each spring have 18 times more DNA than a human does? The idea of a more complex organism needing more DNA seems to break down with some plants. In fact, a lily produces fewer proteins than a human does. Such great amounts of genes are split by long non-coding introns. Only less than 2 per cent of the genome is known to include the exons, the protein coding sequences. You have already seen clusters of VNTRs of variable lengths, arranged in tandem. Further, approximately 1 million copies of short 5 to 8 base pair repeated sequence clustered around the centromeres and near the ends of the chromosomes represent what is called 'the junk DNA'.

Prospects and Implications of Human Genome

It is easy to realize why the genome project is being compared to the discovery of antibiotics. It is expected that we will soon have snapshots of more than 1200 genes that are responsible for common cardiovascular ailments, endocrine diseases like diabetes, neurological disorders like Alzheimer's disease, deadly cancers and many more. Even single gene defects may account for several inherited diseases. Efforts are in progress to determine genes that will revert cancerous cells to normal. The human genome sequencing not only holds promise for a healthier living, it also holds the prospects of vast database of knowledge about designer drugs, genetically modified diets and finally our genetic identity.

15.6 GENE LIBRARIES AND GENE BANKS

You have seen earlier that each chromosome is almost like a volume of a book in a library. Even the genome of a bacterium, such as *E. coli*, contains 4000 genes. Individual genes are present in only very small amounts. As such, gene cloning or more precisely DNA cloning is done to construct several copies of the desired genes in large amounts. This precise technique allows specific DNA sequences to be separated from other sequences permitting detailed analysis or manipulation. **Gene libraries** can also be created using RNA. The enzyme Reverse Transcriptase, a RNA-directed DNA polymerase, is used to convert the RNA into complementary DNA or cDNA, which can then be converted in the same way as DNA cloning. These gene libraries are maintained through special techniques.

A **gene bank** is repository of clones of known DNA fragments, genes, gene maps, seeds, spores, frozen sperms or eggs or embryos. These are stored for possible use in genetic engineering and breeding experiments where species have become extinct. These gene banks will be used increasingly as the rate of extinction increases, depleting the Earth's biodiversity and genetic variety. Now the genomic information is available for a dozen of species. The most remarkable is the prospects and contribution of Human Genome Project.

SUMMARY

Genetic engineering involves the isolation of specific genes,making of copies of genes and their transfer to target genomes. The key to genetic engineering technology is a special class of enzymes called restriction endonucleases, which recognise specific sites and cleave DNA molecules into sticky or blunt fragments. The cleaved two strands of DNA can be joined back together with the aid of a sealing enzyme, ligase. Plasmids and artificial chromosomes from bacteria and yeast are important tools for indirect gene transfer. Bacteria, yeast and cultured plant and animal cells are commonly used as hosts for recombinant DNA experiments. Agriculture has been a major focus of genetic engineering activity. Successes have included incorporation of genes for herbicide resistance into crop plants, conferring natural resistance to insect pests. Genetic technologies have made many advances in the production of affordable drugs including human insulin and interferon. Several recombinant vaccines directed against human diseases such as hepatitis and herpes, are now available as a therapeutic tool in the prevention of disease. In gene therapy, a faulty gene is replaced with a normal healthy gene, to treat diseases where other medical approaches are not effective. With cloning of "Dolly" sheep, the cloning of any mammalian species is now virtually possible. Many useful transgenic microbes, crop plants and farm animals have been cloned successfully. Because the DNA of an individual or species is unique, the genetic analysis methods can be used to identify an organism from a small sample of its cells, that is, to create a DNA fingerprint. It is being effectively applied in forensic science. Genomic libraries can be created as sources of genes for cloning and their use. The blue-print transcript of entire DNA sequence of a human, besides many other comparative genomes, is the most significant achievement of Human Genome Project. Despite benefits, the ethical, social and legal implications of these potent gene technologies have led to considerable public concern over the possibility of accidentally producing new pathogens or developing "genetic monsters".

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EXERCISES

1. Which is the first true example of mammalian cloning?
2. Name at least three therapeutically important products obtained through recombinant genetic engineering.
3. Name at least three diseases against which genetically engineered vaccines are now available.
4. Give one example each of transgenic plant and transgenic animal.
5. What is Gene Therapy? Give at least one example of its application.
6. What is genetically modified food?
7. What are Variable Number Tandem Repeats or VNTRs?
8. Restriction endonucleases
   (a) cleave DNA at highly specific recognition sequences.
   (b) are inserted into bacteria by bacteriophages.
   (c) are made only by eukaryotic cells.
   (d) add methyl groups to specific DNA sequences.
   Choose the correct answer from the above options.
9. Plasmids
   (a) are circular protein molecules.
   (b) are required by bacteria.
   (c) are tiny bacteria.
   (d) confer resistance against antibiotics.
   Choose the correct answer from the above options.

10. Genetic diagnosis by DNA testing
    (a) detects only mutant and normal alleles.
    (b) can be done only on eggs or sperms.
    (c) involves hybridization to ribosomal RNA.
    (d) utilizes restriction enzymes and a polymorphic site.
    Choose the correct answer from the above options.

11. DNA cannot be introduced into any cell by
    (a) microinjection.
    (b) being complexed with calcium salts.
    (c) being placed along with the cell into a gene gun.
    (d) gel electrophoresis.
    Choose the correct answer from the above options.

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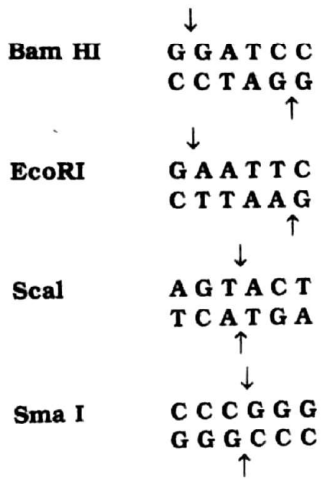
12. In DNA fingerprinting
    (a) a positive identification can be made.
    (b) multiple restriction enzyme digests/generate unique fragments.
    (c) the polymerase chain reaction amplifies fewer DNA.
    (d) the variability of repeated sequences between two restriction sites is evaluated.
    Choose the correct answer from the above options.

13. Here are the recognition sites of restriction enzyme



    Which of these enzymes will create compatible (complementary) sticky end? and which of these will create blunt end?

14. Why is "Agrobacterium-mediated genetic transformation" in plants described as natural genetic engineering of plants?

15. Why is the mother's DNA sample also used in the misidentified paternity cases?

16. Differentiate between
    (i) Cell cloning and organismal cloning.
    (ii) Direct gene transfer and indirect gene transfer.

17. What is a cloned animal? Give one example.

18. What is Southern blotting? Give an example where it is applied?

19. What is genetic engineering? Explain briefly the distinct steps common to all genetic engineering technology.

20. Give an outline of the steps involved in DNA fingerprinting.

21. What is the Human Genome Project? What has been revealed about our genome so far?

CHAPTER 16

MORPHOLOGY OF FLOWERING PLANTS

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The flowering plants also referred to as angiosperms, are the most dominant plants of the present day. Geologically, they are recent, comprising about 300,000 species with a marked diversity in their form and structure. You have learnt that scientists have grouped them into different categories for proper and effective study, where morphology has played an important role. The study of various external features of the plant is known as **plant morphology.** In this Chapter, we will study morphology of angiospermic plants.

These plants show a great variety of shape, size and form. The size ranges from the minute *Wolffia* and *Lemna* (0.1cm) to the tall *Eucalyptus* (upto 100 metre) and large-sized Banyan (*Ficus benghalensis*). In habit, they range from herbs and shrubs to trees.

The life-span of angiospermic plants is highly variable. Plants like pea and gram live only for a few weeks, whereas the famous *Bodhi* tree (*Ficus religiosa*) at Gaya is about 2,500 years old. There is a marked variation in the habitat of these plants. Some may thrive well on land under moderate climatic conditions (mesophytes), and others in water (hydrophytes), in dry conditions (xerophytes), on other plants (epiphytes), on rocks (lithophytes), on sand (psammophytes) or in saline habitats (halophytes). Most of the flowering plants are autotrophic in their mode of nutrition, although there are some which are parasitic (*Cuscuta*), saprophytic (*Monotropa*) or insectivorous (*Utricularia*).

The plant body consists of a main axis which may be branched or unbranched, bearing lateral appendages. This axis is generally divided into an underground part (root) and an aerial part (shoot). These are also referred to as root and shoot systems. The former develops from radicle and is usually brown. The shoot comprises stem, leaves, flowers and fruits. These aerial parts originate from plumule. Flower is a modified shoot and gives rise to fruits and seeds. Seeds develop into new plants. Figure 16.1 shows the sketch of a typical angiosperm.

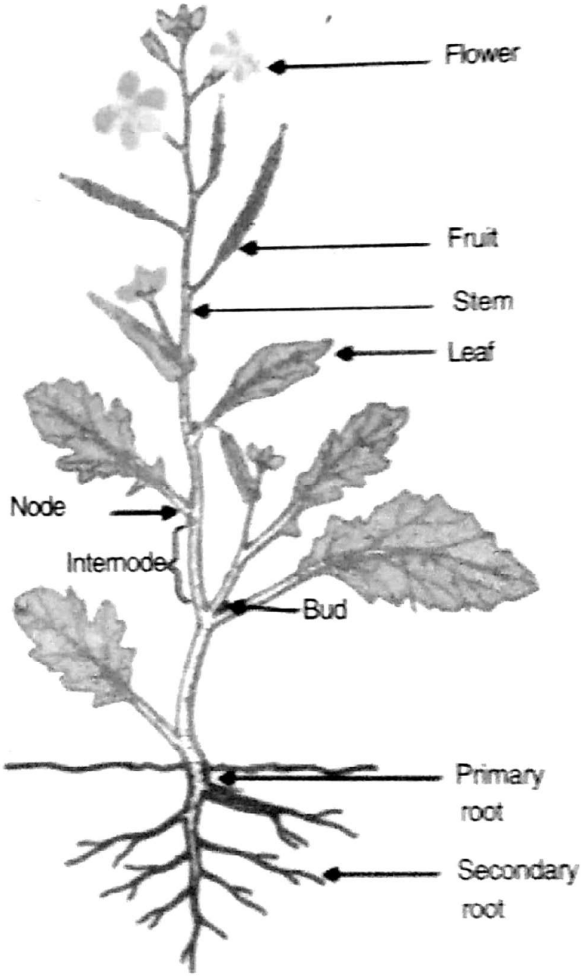

Fig. 16.1 *An angiospermic plant*

16.1 THE ROOT

The root is the underground part of the plant and develops from the elongation of radicle. The first formed root is known as the **primary root**, and others originating from it are secondary or tertiary roots. There are two patterns of growth in roots. In the first, primary root persists as the main root with many secondary and tertiary roots. This is described as the **tap root system** and generally occurs in dicotyledonous plants. In the second, the primary root is short-lived and is replaced by many slender roots, generally uniform in size. They develop from the base of the stem and form **fibrous root system** (Fig. 16.2). These are common in monocotyledonous plants. The main functions of the root system are to absorb water and mineral salts from the soil, and to provide a proper anchorage. Besides these, roots also become modified to perform specialised functions such as storage of food and mechanical support.

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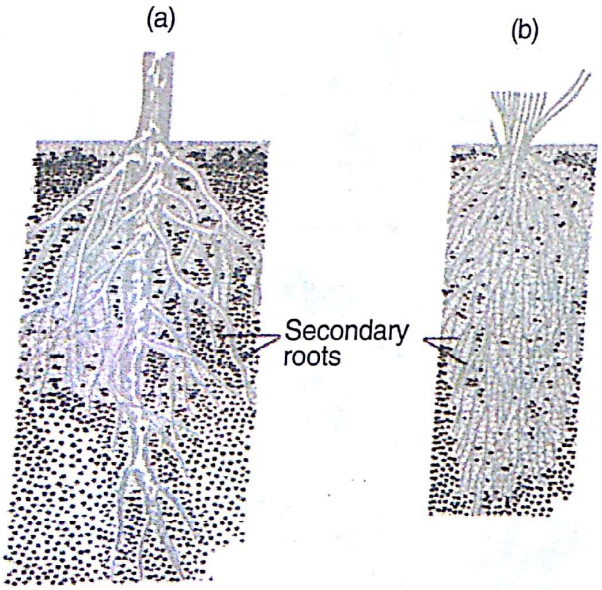

Fig. 16.2 *Root systems (a) tap root system (b) fibrous root system*

Root Zonation

The roots do not bear buds (exceptions, sweet potato and wood-apple), nodes or internodes. The tender apex of the root is protected by a thimble-like structure called **root cap**, which may appear like a loose sheath called **root pocket** in aquatic plants (*Pistia* and *Eichhornia*). Just above the root cap is the region of cell division which extends to a few millimetres. The cells of this region are small, thin-walled with dense protoplasm, and undergo repeated divisions. This is also referred to as **meristematic region.** The **region of elongation** lies above the meristematic region and is slightly longer. The cells of this region undergo rapid elongation and enlargement and are responsible for growth of the root in length. The **region of maturation** is above the region of elongation and extends upwards (Fig. 16.3). The cells of this region undergo maturation and differentiation into various tissues. A cluster of fine, delicate thread like structures called root hair, emerge from the region of maturation.

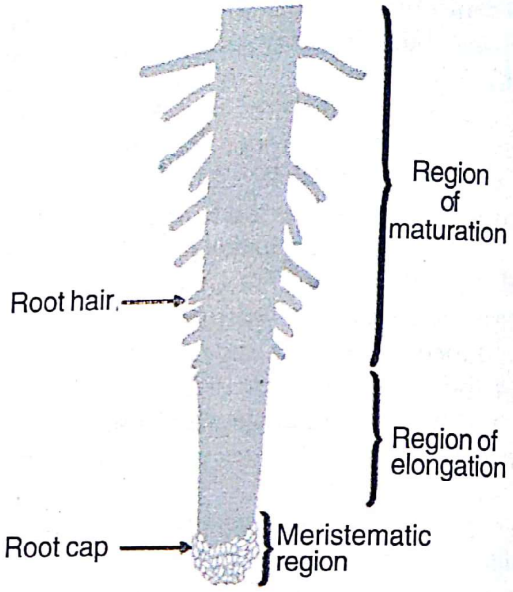

Fig. 16.3 *Regions of root tip*

Adventitious Roots

Roots developing from any part of the plant other than radicle are called adventitious roots. These roots replace primary root or are in addition to it or develop from nodal/internodal regions of the stem or even from leaf. They perform both normal and specialised functions of the roots. In monocotyledons, they develop in clusters from the base of the stem as in onion or from nodes of branches as in grasses. In some cases the roots are produced spontaneously whereas in others they may develop from the petiole or veins of the leaf due to some injury. These are called **foliar** roots and can also be induced by the application of hormones. Several plants creeping on the ground produce roots from nodes as in *Oxalis* or from branch

cuttings when these are put into the soil as in rose, sugarcane, tapioca or when kept partially immersed in water as in *Coleus*. Some foliar buds also produce adventitious roots as in *Bryophyllum* and *Begonia*.

Modifications of Root

The excess food synthesised by the leaves in some plants needs to be stored elsewhere. The primary root of radish stores this excess food and gets swollen in the middle and tapers towards the apex and base, giving it the appearance of a spindle. Such a root is called **fusiform** root (Fig. 16.4a). In turnip and beet, the root gets swollen at the upper part, becoming almost spherical and abruptly tapering at the lower part. It is termed as **napiform** root (Fig. 16.4b). If the root is broad at the base and tapers gradually towards the apex, it is called **conical** root as in carrot (Fig. 16.4c). The root, when thick and fleshy without any definite shape as in *Mirabilis*, is of **tuberous** type (Fig. 16.4d).

Sometimes the secondary and tertiary branches of the tap root and even adventitious roots, become modified for the storage purpose. For example, in sweet potato the roots are swollen without any definite shape. These are **tubercular** roots. In *Dahlia* and *Asparagus* many tubercular roots develop in clusters or fascicles at the base of the stem. In turmeric, the slender roots are swollen at the apex, giving the form of **nodules**. The swellings at frequent intervals as in *Portulaca* and *Momordica*, give beaded or **moniliform** shape to roots. If the swellings are in a series of rings on the roots, these are called **annulated** as in *Ipomoea*.

In plants like banyan and screwpine, a number of roots are produced from the main stem or branches to provide **mechanical support** to the shoot system. These roots grow downwards, penetrate into the soil and act as supporting pillars. Such roots are known as **prop** roots (Fig. 16.5). Plants like betel, black-pepper having weaker stems, produce roots from their nodes so as to support the plant in climbing on the nearby objects. Some of the stout roots around the base of the main trunk of a tree show prolific abnormal growth and look like planks. These roots give support to huge trunks as in silk cotton tree and kapok.

In some plants, the roots get modified to perform vital functions of the plants like respiration, absorption of water from the atmosphere and even synthesis of food. Plants like *Rhizophora* and *Heritiera* growing in marshy places and saline lakes, develop conical structure (**pneumatophores**) from underground roots which rise vertically upwards above the ground (Fig. 16.6). These possess numerous pores and are called respiratory roots. Parasitic plants, generally non-green, like *Cuscuta* and *orobanche* grow on other plants and suck the food with the help of **haustoria** or sucking roots. Some orchids like *Vanda* grow on the branches of trees and develop **aerial** roots with spongy tissue (velamen). This tissue helps in the absorption of moisture from the surrounding air. The long, slender hanging roots in *Tinospora* and some orchids develop chlorophyll and carry out photosynthesis. Such assimilatory roots are also developed in water chest-nut.

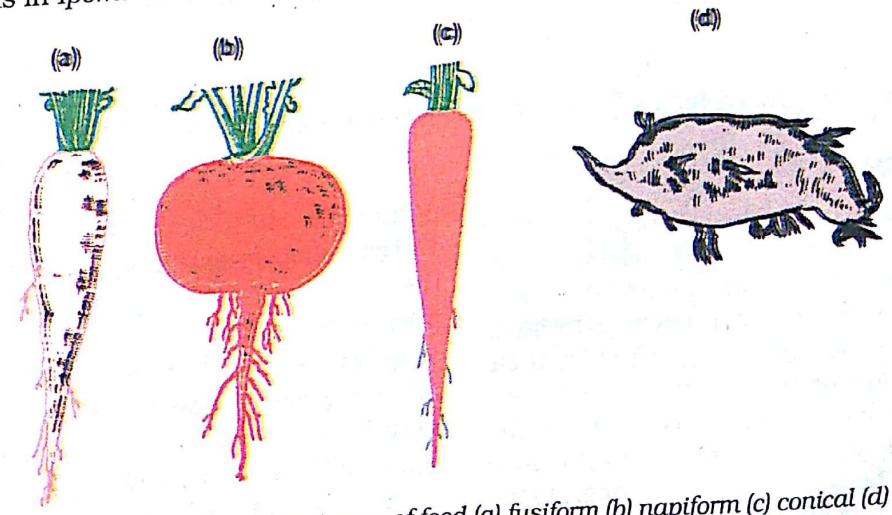

Fig. 16.4 *Modifications of root for storage of food (a) fusiform (b) napiform (c) conical (d) tubercular*

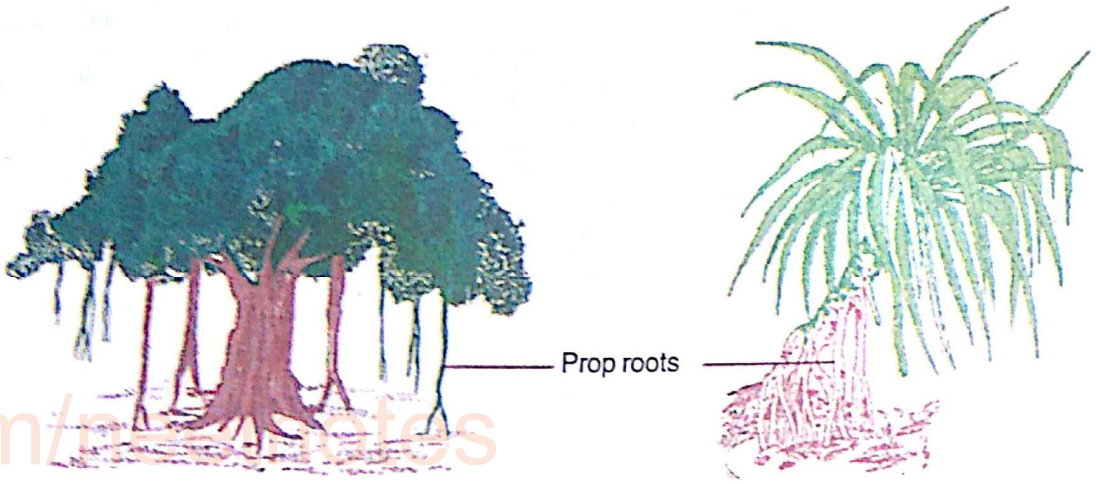

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Fig. 16.5 Modifications of root for mechanical support

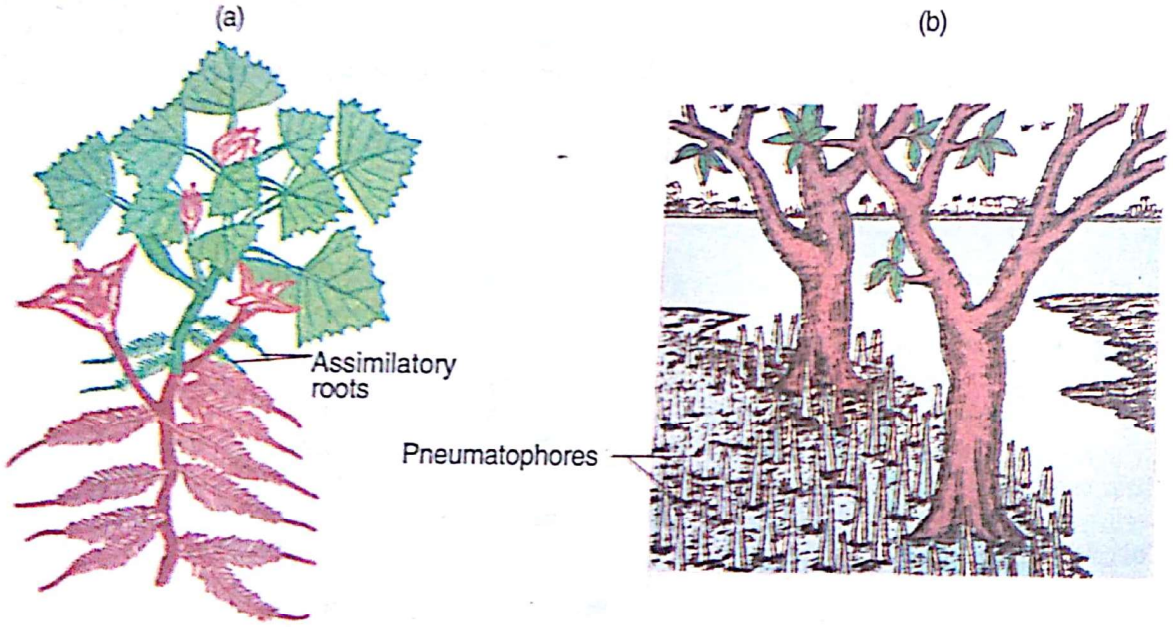

Fig. 16.6 Modifications of root for vital functions (a) assimilatory roots (b) pneumatophores

16.2 THE STEM

Stem, developed from the plumule, is the ascending part of the plant axis. Generally, it grows above the ground and bears branches, leaves and flowers. The main function of the stem is to properly spread out these organs for their specific functions (manufacture of food and production of fruits/seeds). It also helps in the conduction of water, mineral salts and photosynthates, besides providing support to the branches. In addition to the positive phototropic nature, the presence of nodes and internodes and exogenously developed multicellular hair, are the chief characteristics of the stem. The growing apex of the stem is generally a dome-shaped structure covered and protected by a number of minute leaves. The stem also bears buds, which may be terminal, axillary or accessory and protected by scales. Stem is generally green and herbaceous in early stages, and may later become woody and dark-brown in colour.

Forms of Stem

The stem is adapted to perform a variety of functions under different conditions. It may be erect, rigid, strong and remain upright. In some cases it may be unable to support itself in the upright position and may trail on the ground or climb on neighbouring objects (Fig. 16.7). In still others, it is underground and produces aerial branches under favourable conditions.

If the stem is unbranched, erect, cylindrical and stout, and marked with scars and remnants of fallen leaves as in palms, it is called **caudex**. In bamboos the stem is jointed with solid nodes and hollow internodes and is known as **culm**. In some other monocotyledons there is no aerial stem, instead there are aerial shoots which bear flowers. These shoots are termed as **scape,** e.g. onion and aroids. If the stem trails on the ground and lie prostrate (*Oxalis* and *Evolvulus*), it is called **prostrate** or **procumbent**. In *Tridax* the stem trails for some distance and then tends to rise at its apex; it is termed as **decumbent**. When the branches of the stem are spread out in all directions on the ground as in *Boerhaavia* it is said to be **diffuse.** The **climbers** are those stems which attach themselves to nearby object by means of some special devices like hooks as in *Bougainvillea* or tendrils as in wild pea.

Modifications of Stem

In some plants, the stem gets modified into different types of structures. These modifications help the plant to survive during unfavourable seasons by storing food, by propagating vegetatively and by providing mechanical support and protection. Various modifications of stems can be studied by grouping them into underground, subaerial and aerial types.

Underground Modifications

The stems of some plants may remain underground permanently, generally in a dormant state, and produce aerial shoots annually under favourable conditions. They contain sufficient amount of reserve food materials and bear roots and buds. Such stems can be used as 'seeds' to produce new plant (Fig. 16.8). A prostrate thickened stem with distinct nodes, internodes, buds and scale leaves, which creeps horizontally under the ground is a **rhizome**. The examples of this type of stem are ginger, turmeric, canna, water lily, some ferns and many aroids. **Corm** is a condensed form of rhizome growing more or less in vertical direction with axillary buds and scale leaves. It is found in zamikand, colacasia and saffron. An underground branch of the stem which develops from the axil of lower leaf, grows horizontally outward and swells at the apex, is **tuber.** It has a number of buds (eyes) which grow into new plants as in potato. **Bulb** is a modification of complete shoot and consists of shortened convex or slightly conical stem, terminal bud and many scale leaves. Clusters of fibrous roots develop from its base. The fleshy scale leaves store food as in onion and garlic.

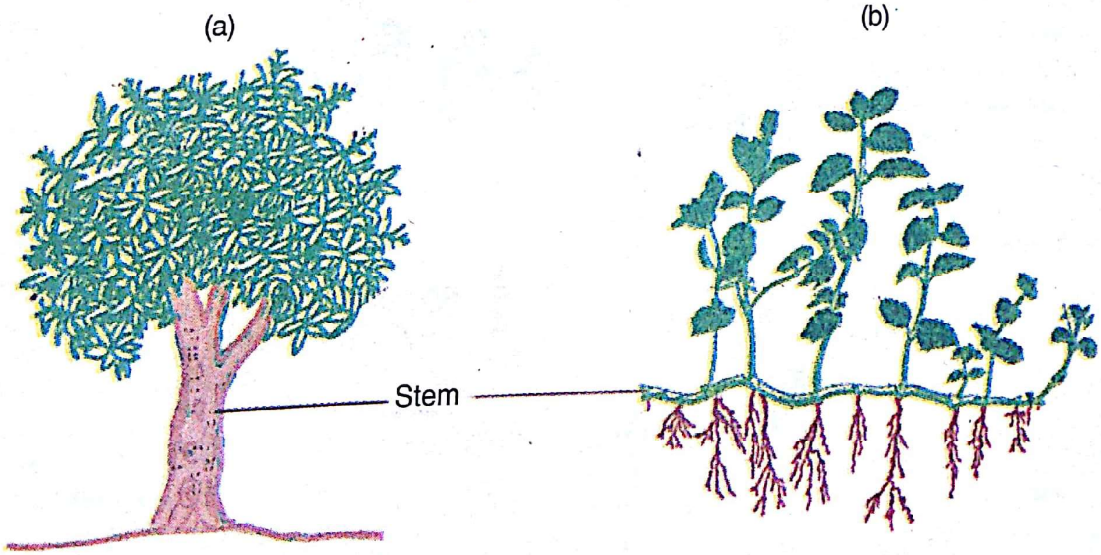

Fig. 16.7 *Forms of stem (a) erect, strong stem (b) trailing stem*

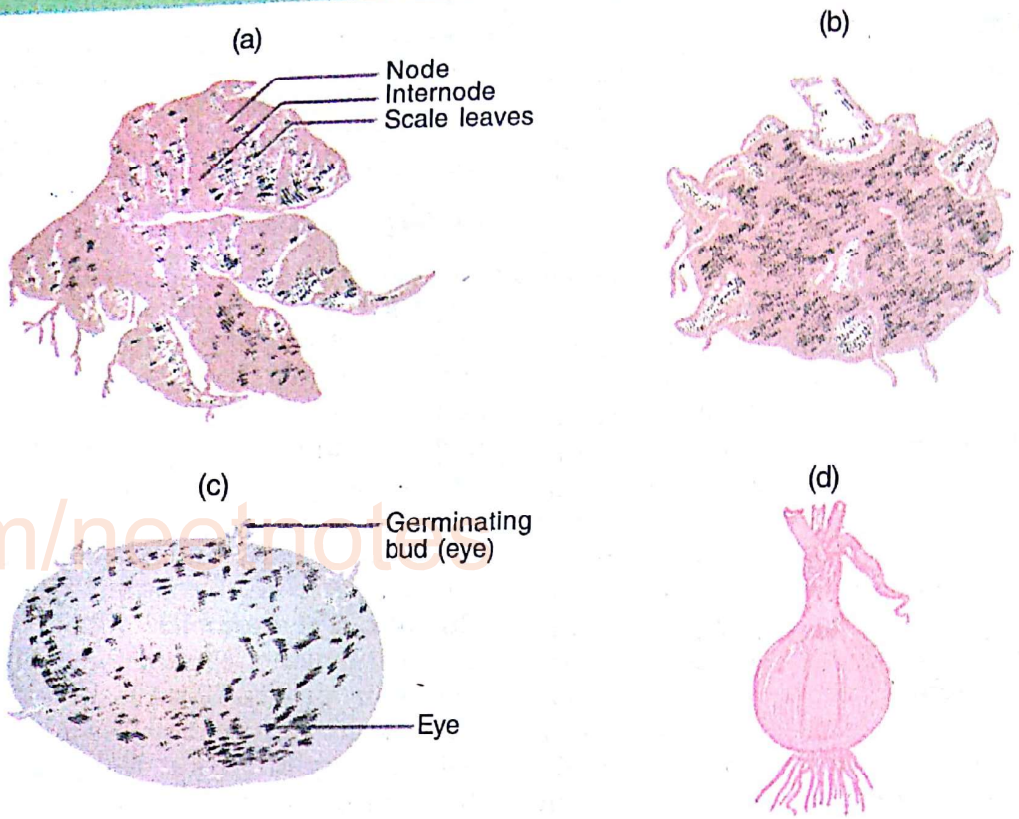

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Fig. 16.8 *Underground modifications of stem (a) rhizome (b) corm (c) tuber (d) bulb*

Subaerial Modifications

In some species, the stem is partly aerial and partly underground (Fig. 16.9). **Runner** includes a slender, prostrate branch arising from an axillary bud and creeping on the ground with roots at nodes. It may break off from mother plant and grow independently as in *Oxalis*. In plants like wild strawberry and peppermint, the

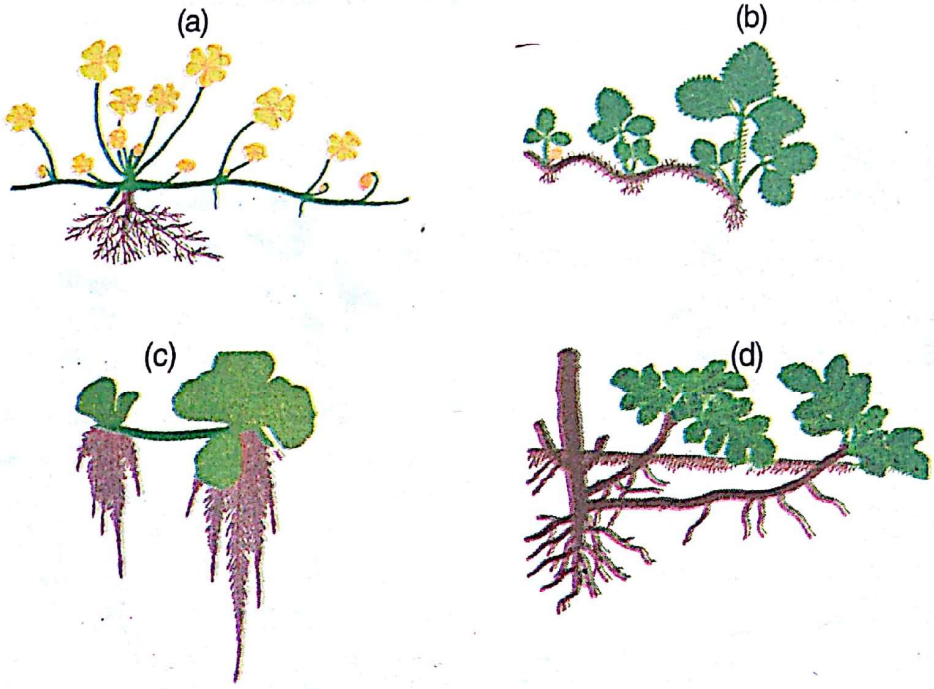

Fig. 16.9 *Subaerial modifications of stem (a) runner (b) stolon (c) offset (d) sucker*

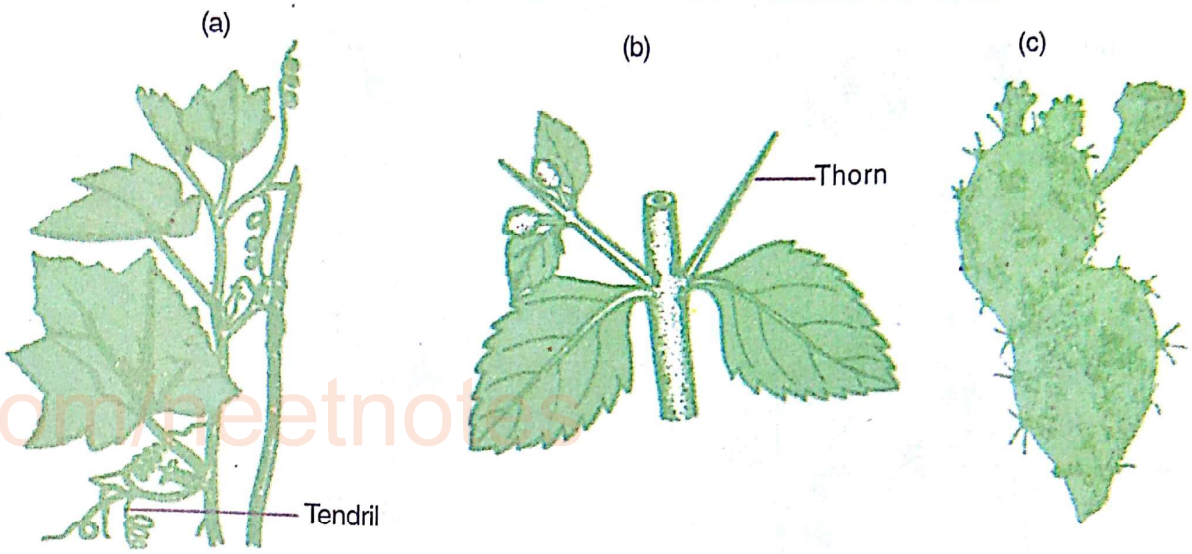

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Fig. 16.10 Aerial modifications of stem (a) tendril (b) thorn (c) phylloclade

branches originate from the base of the stem and grow obliquely. These are referred to as **stolon**. **Offset** originates in the axil of leaf as a short, thickened horizontal branch, which elongates to produce a tuft of leaves above and clusters of small roots below, e.g. *Pistia* and *Eichhornia*. In banana, pine-apple and *Chrysanthemum*, the laterally developed branches grow obliquely upward and give rise to leafy shoots. They are **suckers** and produce new plant after getting separated from the mother plant.

Aerial Modifications

The stem or its parts may become modified into **tendrils,** to provide support and help the plant in climbing as in passion-flower and *Vitis*. In lemon, pomegranate and karonda, axillary and terminal buds of the stem are modified into **thorns** and give protection to the plant. The **prickles** are also modifications of stem and act as climbing organs. They develop superficially at the surface of stem and are distributed irregularly as in rose and plum. **Phylloclades** are green, flattened or cylindrical branches of unlimited growth. These occur in xerophytic plants like *Opuntia*, *Casuarina* and some species of *Euphorbia*. In these, the leaves may fall early or get modified into spine. In *Asparagus*, the branches of limited growth become green and flat like a leaf. These are called **cladodes**. **Bulbil** is a modified vegetative bud with stored food meant for reproduction, as in *Agave*. Figure 16.10 illustrates some of the aerial modifications of stems.

Branching of Stem

The main axis (stem) of the shoot produces branches, which arise either laterally or dichotomously (Fig. 16.11). The lateral branching is from the side of the main axis and may have limited growth (cymose type) or unlimited growth (racemose type). **Cymose** type of branching is from terminal buds and the branches stop growing after some time. The lateral branches grow more vigorously, and allow the plant to spread into more or less a dome. If only one lateral branch is produced at a time, the system is **uniparous** or **monochasial** as in *Saraca* (one-side) or vine (alternate sides). In *Datura* and *Mirabilis*, two lateral axes develop at the same time and the branching is **biparous**. If more than two branches develop at a time, the branching is **multiparous** as in some species of *Croton* and *Euphorbia*. The **racemose** branches continue to grow indefinitely and give off further branches laterally in an acropetal order. As a result, the plant takes a conical or pyramidal outlook as in *Polyalthia* and *Casuarina*. In some, the terminal bud bifurcates producing two branches in a forked manner. Screwpine and *Hyphaene* (a kind of palm) are examples of this category.

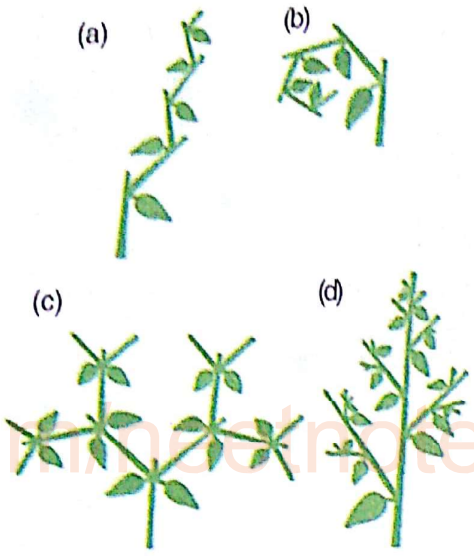

Fig. 16.11 *Branching of stem (a, b) uniparous cyme (c) biparous (d) racemose*

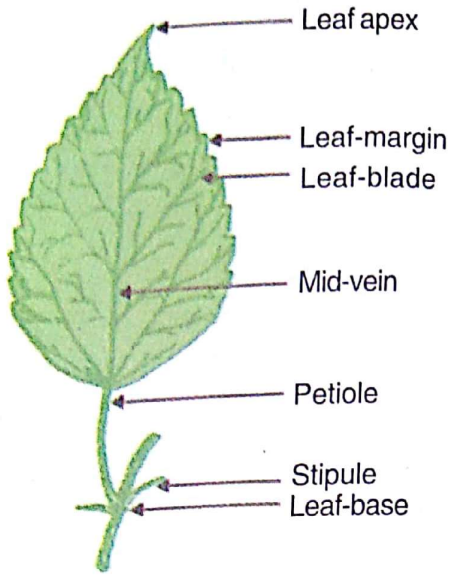

Fig. 16.12 *Parts of a leaf*

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16.3 THE LEAF

A leaf is the lateral appendage of the stem or its branch, and develops at the node. Leaves are arranged in an acropetal order and originate from leaf primordia. Being rich in chlorophyll and green in colour, they constitute the most conspicuous vegetative part of the plant. The duration of leaf on the plant body varies. If these fall soon after appearance or just after the opening of bud, they are **caducous**. Those which continue to remain attached to the stem for one season, usually falling off in winter, are **deciduous**. The leaves persisting for more than one season, usually lasting for a number of years, are **persistent.**

Parts of a Leaf

A typical leaf consists of three main parts, namely leaf base, petiole and lamina (Fig. 16.12). The **leaf base** is attached to the stem and in many cases it bears lateral leafy outgrowth (stipule). In some monocot plants, it expands into a sheath, covering the stem partially or wholly. In many leguminous plants, the leaf base may become swollen and is known as **pulvinus**. The **petiole** or stalk is the other part of the leaf that pushes the leaf-blade to secure proper sunlight. In some plants, it may be absent and thus, the leaves are sessile. Generally, the petiole is cylindrical, smooth or groved, but in *Eichhornia*, it swells and in *Citrus*, it is winged. In Australian acacia, it is modified into a sickle-shaped lamina - a leaf like structure (phyllodes).

The **leaf-blade** or lamina is the green, expanded part of the leaf with veins and veinlets. There is a marked variation in features, such as shape, margin, apex, surface and extent of incision (Fig. 16.13). It may be needle-shaped as in pine, linear as in grasses, lanceolate as in bamboo and *Nerium*; oval as in guava and *jamun*; ovate as in china-rose and banyan; oblong as in banana; orbicular as in lotus, cordate as in betel; oblique as in *Begonia* and *neem*; spathulate as in sundew and calendula; sagittate as in aroids; falcate as in eucalyptus; lyrate as in radish. The margin of the lamina may be entire, wavy, deeply undulating, serrate, dentate, spinous, etc. The leaf apex may be obtuse (banyan), acute (china rose), acuminate (peepal), cuspidate (date-palm), ratuse (*Pistia*), emarginate (*Bauhinia*), cirrhose (banana), etc. The surface may be smooth, rough, spiny, hairy, etc.

Venation of the leaf-blade helps in conduction of water, mineral salts, and prepared food, besides providing strength to the lamina. Generally, it is **reticulate** in dicot plants and **parallel** in monocot plants. It may be unicostate reticulate as in the leaves of mango and peepal, unicostate parallel as in banana and canna; multicostate reticulate as in castor, china rose and plum, and multicostate parallel as in

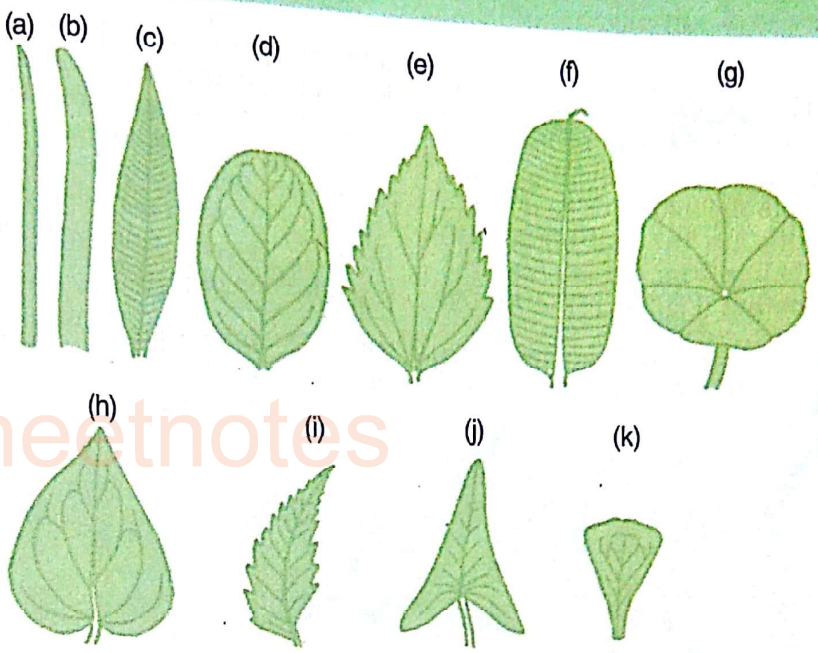

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Fig. 16.13 *Types of leaf-blade (a) needle-shaped (b) linear (c) lanceolate (d) oval (e) ovate (f) oblong (g) orbicular (h) cordate (i) oblique (j) sagittate (k) spathulate*

grasses and palms (Fig. 16.14). The incision of the leaf-blade may proceed from margin towards the midrib (pinnate type) as in cosmos, *Cassia* or may proceed towards the base (palmate type) as in castor, tapioca and silk cotton tree (semal).

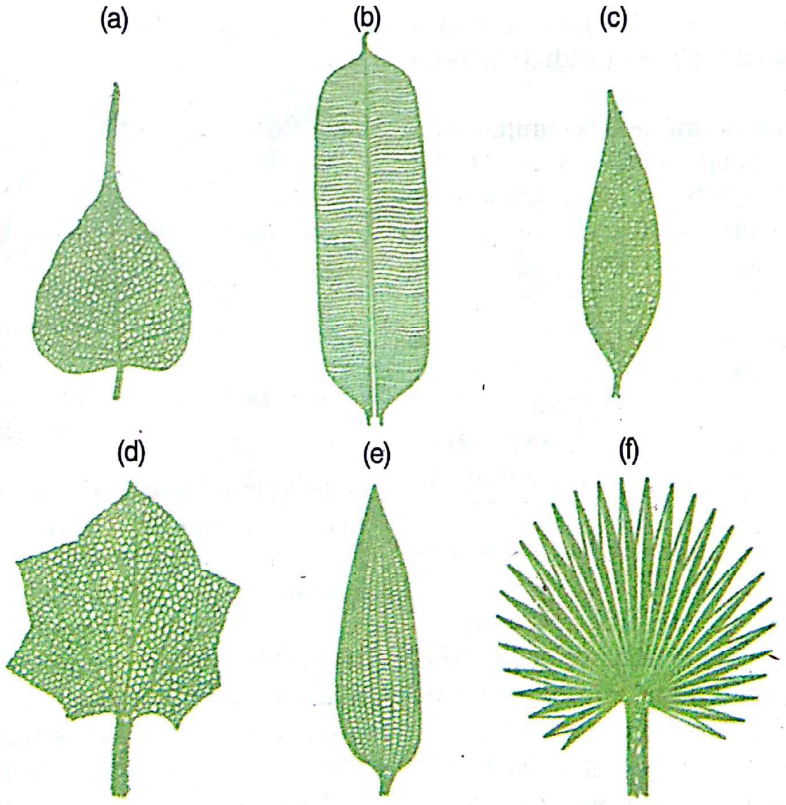

Fig. 16.14 *Venations in leaf (a) unicostate reticulate (b) unicostate parallel (c, d) multicostate reticulate (e, f) multicostate parallel*

Types of Leaf

A leaf is said to be **simple**, if it consists of a single leaf-blade. The blade may be entire or incised to a variable degree of depth. If the incisions reach upto the midrib or go down to the petiole breaking up the leaf-blade into segments or leaflets free from one another, the leaf is called **compound** (Fig. 16.15). A bud is present in the axil of petiole in both simple and compound leaves, but not in leaflets of the compound leaf. If the rachis or midrib of a compound leaf bears a number of laterally present leaflets, the leaf is said to be **pinnate** type. In **palmate** type, the petiole bears terminally articulated leaflets or radiating lobes as in silk cotton tree.

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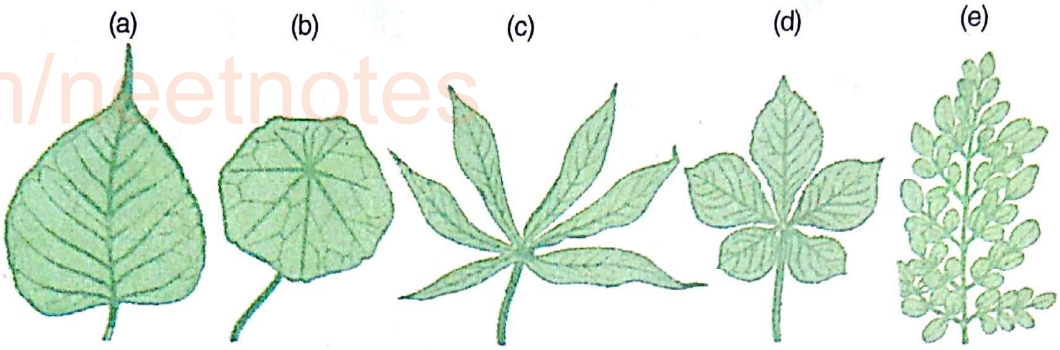

Fig. 16.15 *Types of leaf (a-c) simple (d, e) compound*

The pinnately compound leaf is unipinnate, if it bears leaflets directly on the rachis. If the leaflets are even in number it is paripinnate as in *Cassia* and *Sesbania*, and if odd in number, it is imparipinnate, as in neem and rose. In *Acacia* and *Mimosa*, the leaf is bipinnate and the leaflets present on secondary axes are produced by the midrib. In tripinnate leaves the leaflets are produced on tertiary axes as in *Moringa*. The leaf is decompound, if it is pinnate more than thrice as in coriander and carrot. The palmately compound leaves may be unifoliate, bifoliate, trifoliate and so on, depending on the number of leaflets present.

Leaves are termed as foliage, if they are green, flat, borne laterally at node of stem or branch. They are cotyledonary, if attached to the axis of embryo, and scaly, if reduced, stalkless, generally brownish as those present on buds and rhizomes. The bracts are also leaves, developed at the axil of the flower or inflorescence. The stipules appear as the lateral appendages of the leaf, borne at its base, whereas ligules are minute scaly outgrowths borne at the upper end of the leaf-sheath as in grasses. The floral leaves like sepals and petals are the non-essential parts of the flowers, whereas sporophylls are spore-bearing leaves and are concerned with reproduction of plants.

Phyllotaxy

Phyllotaxy is the pattern of attachment or arrangement of leaves on the stem or branch (Fig. 16.16). When a single leaf arises at each node as in mustard, china rose, sunflower, it is said to be of alternate type. In several plants, two leaves arise from each node generally opposite to each other. Each successive pair of leaves stands at right angles to the next one in decussate opposite condition as in *Calotropis*. When the pair of leaves stands directly over the lower pair, it is called **superposed** type as in guava. Sometimes there are more than two leaves at each node arranged in a **whorl** as in *Alstonia* and *Nerium*.

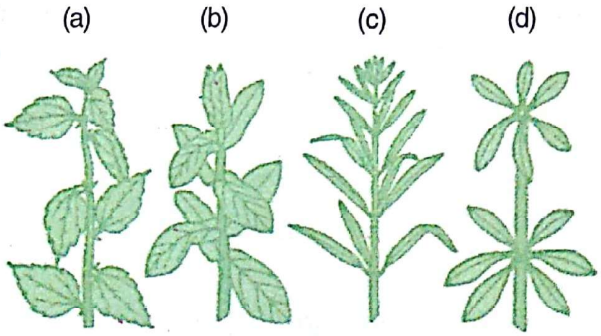

Fig. 16.16 *Types of phyllotaxy (a) alternate (b) opposite (c, d) whorled*

Modifications of Leaf

Although the major function of leaves is to synthesise food for the plant, in some cases they get modified into distinct forms to perform the functions of support and protection to plant, and storage of food (Fig. 16.17). For example, in sweet and wild peas, and glory lily, the leaves are modified into slender, wiry, closely coiled structures called **tendrils**. These are sensitive and work as climbing organs for the plant. In cat's nail (*Bignonia unguis-cati*), the terminal leaflets are modified into curved **hooks** for helping the plant in climbing. In *Argemone*, *Opuntia* and *Aloe*, the leaves get modified into sharp, pointed structures (spines) for defensive purposes. Sometimes the leaves become thin, dry, stalkless, membranous structures and serve to protect axillary buds as in *Ficus* and *Tamarix* or store food and water as in onion. In Australian acacia, the petiole becomes flat, green and leaf like, the **phyllode**. The leaves in some plants are modified into a **pitcher** with a lid to catch insects as in *Nepenthes* or into **bladder** as in *Utricularia*.

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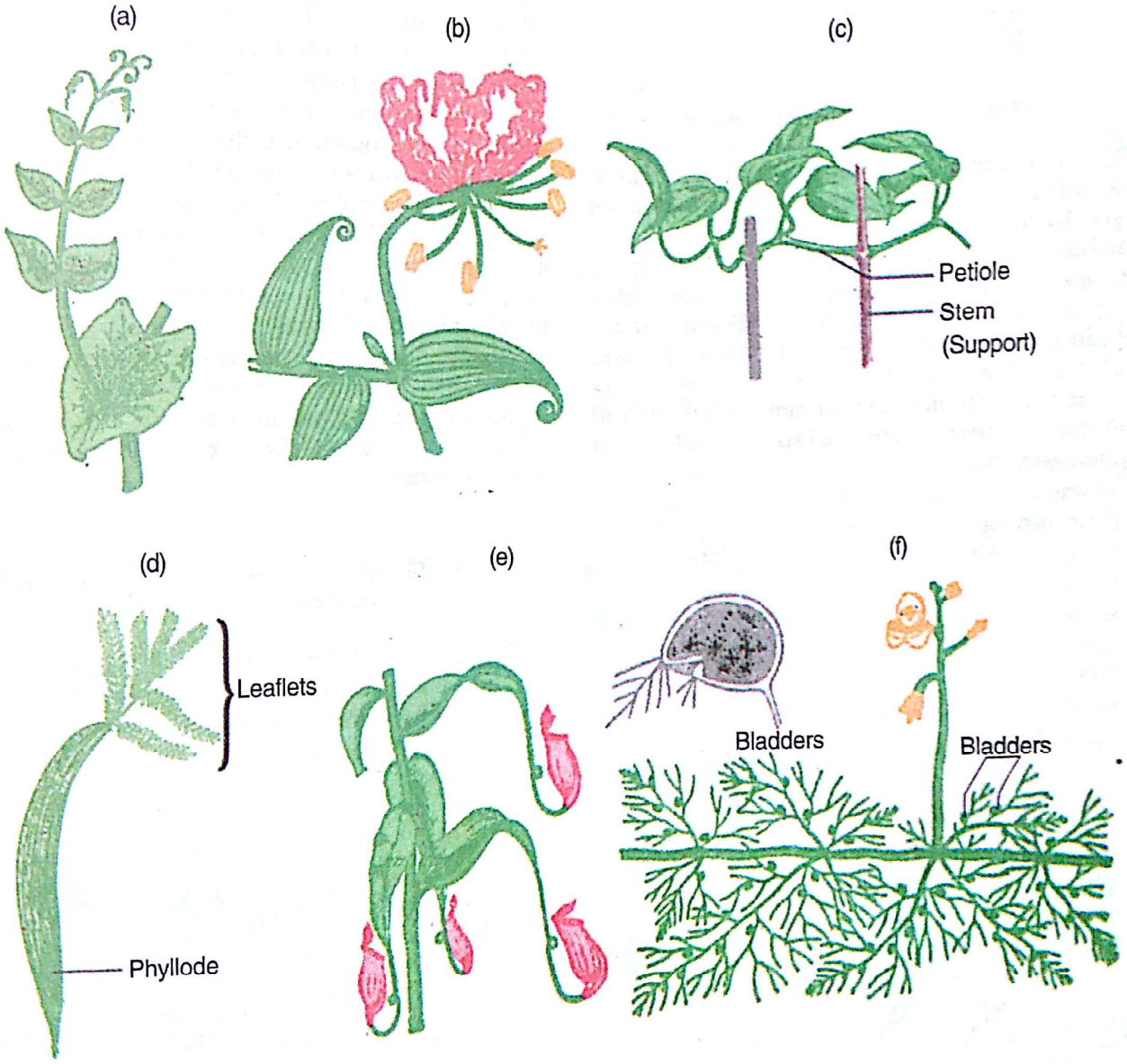

Fig. 16.17 Modifications of leaf (a, b) tendrils (c) twinging petiole (d) phyllode (e) pitcher (f) bladder

Heterophylly

Some plants show more than one type of leaves and this phenomenon is called heterophylly (Fig. 16.18).

Fig. 16.18 Heterophylly in plants

This is common in aquatic plants growing in running water. The floating or aerial leaves are broad, fully expanded, whereas the submerged leaves are narrow, ribbon-shaped, linear or highly dissected. *Ranunculus aquatilis*, and some species of *Sagittaria* are examples of this feature. However, some land plants like *Artocarpus heterophyllus* (jackfruit), *Hemiphragma heterophyllum* and *Ficus heterophylla* also exhibit this phenomenon.

16.4 THE INFLORESCENCE

Inflorescence is the arrangement of flowers on the floral axis. The stalk bearing an inflorescence is called peduncle. It arises terminally or in axil and may have a number of flowers. The peduncle may be branched in various ways. The inflorescences are of various types depending on the type of branching of the peduncle. There are two major types – namely **racemose** with indefinite growth, and **cymose** with definite growth of the main axis.

In the racemose type of inflorescence, the main axis does not terminate into a flower, but continues to produce flowers laterally in an acropetal order. The elongated main axis, bearing a number of stalked (pedicellate) flowers laterally as in radish, mustard and dwarf gulmohar represent the raceme inflorescence. In *Adhatoda* and amaranth, the sessile flowers are arranged on the elongated main axis which is referred to as **spike**. In the members of grass family, the small spikes with one or a few flowers are called **spikelets**. In mulberry, *Betula* and oak, the spike with long and pendulous axis bears unisexual flowers only. This is termed as **catkin**. The spike with fleshy axis enclosed by brightly coloured bracts (spathe) as in banana, aroids and palms, is **spadix**. Fig. 16.19 illustrates some of the racemose types of inflorescence.

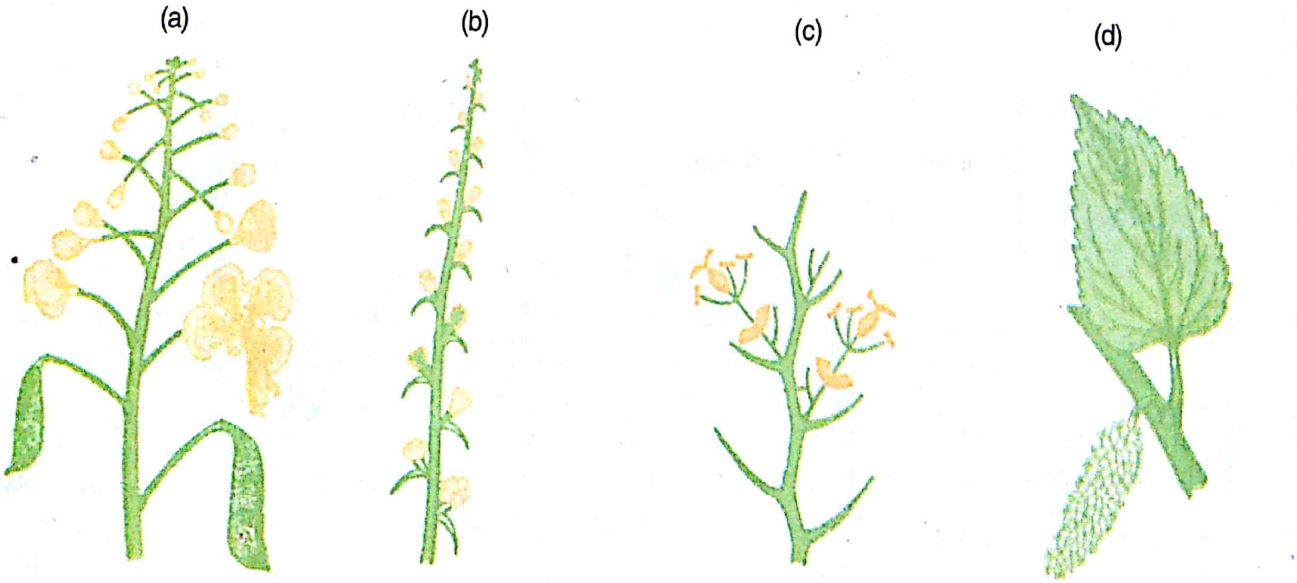

Fig. 16.19 Types of racemose inflorescence (a) raceme (b) spike (c) spikelet (d) catkin

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In **corymb** inflorescence, the main axis is comparatively short with longer stalks of lower flowers. All the flowers are brought to more or less same level as in candytuft. In **umbel**, the primary axis is shortened and bears a cluster of flowers with a whorl of bracts in the form of involucre as in the members of coriander family. In some inflorescences, the main axis is flattened or even suppressed and bears a group of sessile, very small flowers or florets. Generally, all florets are attached to a flat surface of receptacle and can be divided into marginal (ray florets) and central (disc florets). The complete inflorescence looks like a single flower and is called racemose **head** or **capitulum** as in sunflower, *Zinnia* and marigold. If the main axis is branched and flowers are borne on branches, the inflorescence is **compound.** Compound raceme is found in gulmohur and neem, compound spike in wheat, compound spadix in palms, compound corymb in candytuft, and compound umbel in coriander.

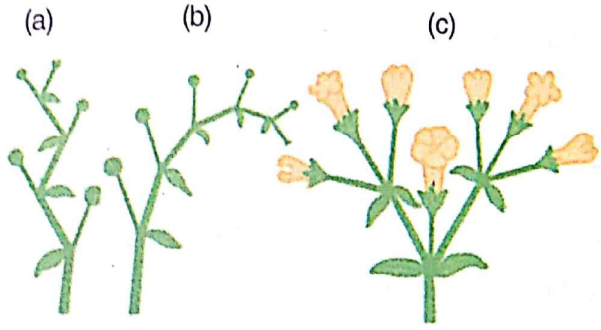

Fig. 16.20 *Types of cymose inflorescence (a, b) uniparous (c) biparous*

In cymose inflorescence (Fig. 16.20), the growth of main axis is checked by a flower at the apex. In **uniparous** or **monochasial** cyme, the main axis ends into a flower and has only one lateral branch, also terminating into a flower as in *Begonia*, sundew, and some species of *Solanum*. In *Bougainvillea*, jasmine and teak, the main axis ends up in a flower and has two lateral branches each with a flower at their ends. It is called **biparous** or **dichasial cyme**. In **multiparous cyme**, the main axis ends in a flower with a number of lateral flowers around it, as in madar and *Asclepias*.

Cyathium is a special type of inflorescence found in *Euphorbia* species. It has a cup-shaped involucre with a single female flower surrounded by several male flowers. A nectar-secreting gland is present at the rim of the involucre.

Verticillaster is a condensed form of cymose with a cluster of sessile flowers in the axil of a leaf, forming a false whorl at the node as in *Salvia*, *Ocimum* and *Coleus*. In **hypanthodium**, a hollow cavity is formed in a pear-shaped receptacle with a narrow apical opening surrounded by scales. The flowers are produced on the inner wall of the cavity as in *Ficus* species. Fig. 16.21 illustrates the special types of inflorescence.

16.5 THE FLOWER

The flower is a modified shoot, meant for sexual reproduction. A typical or complete flower has four main parts namely, **calyx** and **corolla** (accessory organs) and **androecium** and **gynoecium** (reproductive organs). All the four parts are arranged in a cyclic manner on the thalamus - a swollen end of the stalk or pedicel of the flower. A flower with only one reproductive organ either stamens or carpels is unisexual and that with both these organs is bisexual or

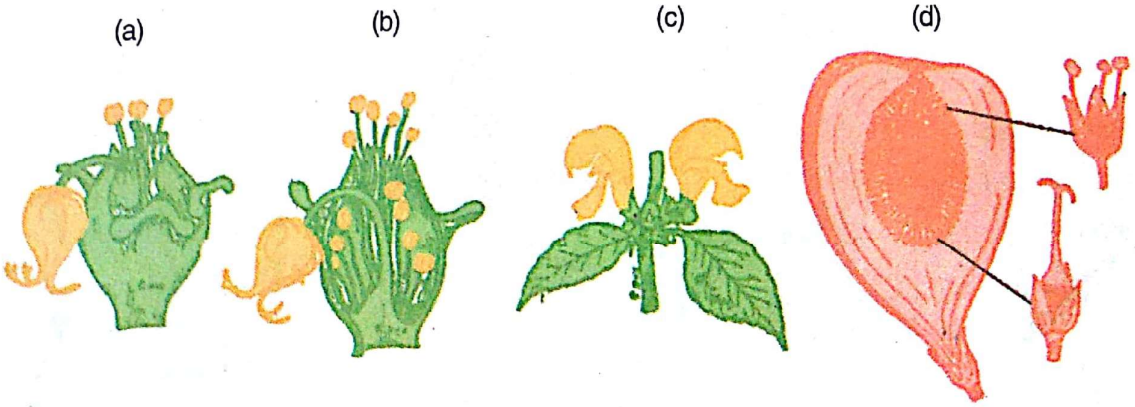

Fig. 16.21 *Special types of inflorescence (a, b) L.S. of cyathium (c) verticillaster (d) L.S. of hypanthodium*

hermaphrodite. Unisexual flowers are said to be imperfect in contrast to perfect flowers which contain both androecium and gynoecium. A plant with unisexual flowers may be either **monoecious,** i.e. both male and female flowers are borne on the same plant as in cucurbits, or **dioecious,** i.e. male and female flowers on different plants as in papaya and mulberry. A plant bearing bisexual, unisexual and even neutral flowers is said to be **polygamous**, e.g. *Polygonum* and mango.

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The flower may be **regular** or **actinomorphic**, if it can be divided into two equal halves through any vertical plane passing through the centre, e.g. mustard, datura, chilli, etc. When a flower divides into two similar halves only in one particular vertical plane, it is **zygomorphic** or **bilateral** as in pea, bean, gulmohur and *Cassia.*

A flower is **asymmetrical** or **irregular**, if it cannot be divided into two similar halves in any vertical plane as in *Canna.* A flower is **isomerous,** when each whorl has an equal number of parts or its multiple. Isomerous flowers may be bi-, tri, tetra- or even pentamerous according to the number of parts in each whorl. Opposite to isomerous is **heteromerous.**

All the floral leaves are inserted on the thalamus. Generally, the thalamus is short and flattened but in some instances, it becomes elongated and shows distinct nodes and internodes. Normally the floral appendages lie on the thalamus in their proper sequence, but in some cases, the relative position of calyx, corolla and androecium in respect of gynoecium becomes disturbed due to unusual growth of the thalamus. These are of three distinct types – Hypogynous, Perigynous and Epigynous. In a **hypogynous** flower, the thalamus is conical, and the gynoecium occupies the highest position on the thalamus, while other parts are present below it. The ovary thus becomes **superior** as compared to other parts as in mustard, china rose and brinjal. In the **perigynous** flower, the thalamus is disc like on which the carpels are borne in the centre and the rest of the floral whorls are located on the rim of the thalamus. The ovary here is said to be half-inferior as in plum and peach. In the epigynons condition the margin of the thalamus grows further upward enclosing the ovary completely and getting fused with it. The sepals, petals and stamens thus arise above the ovary. In this situation the ovary is **inferior,** e.g. sunflower, guava, cucumber and apple. A special type of leaf called bract may be present at the base of the flower. If the leaves are present on the pedicel, they are called bracteole. Flowers with bracts are called **bracteate** and those without bracts **ebracteate**. The axis or part of shoot on which the flower is borne is described as the **mother axis.** The part of flower towards the mother axis is posterior part and the one opposite to it is the anterior part. The region on the right and left side is called the lateral portion. This orientation becomes a necessity when we illustrate the entire arrangement of the floral appendages in the form of a floral diagram.

Parts of a Flower

As mentioned earlier, a typical flower is made up of four parts, namely calyx, corolla, androecium and gynoecium (Fig. 16.22). The calyx is the outermost part and consists of sepals. Generally, the sepals are green but in several instances, they may have colour of the

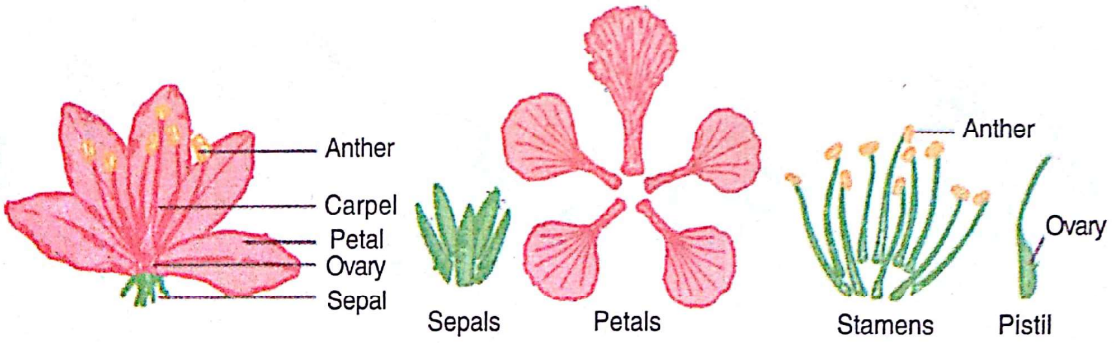

Fig. 16.22 *Parts of a flower*

petals (**petaloid**). They may be united with each other (gamosepalous) or free (polysepalous). In some cases, there may be another whorl of small and green appendages just below the sepals, called **epicalyx** as in china rose. The calyx may be caducous or deciduous, or even persistent depending upon the duration of its retention on the thalamus.

Petals are the individual units of the corolla and occur inner to sepals. The petals may be free or united and regular or irregular in their symmetry. The petals with regular symmetry and polypetalous condition (free) may have different shapes like cruciform, caryophyllaceous and rosaceous. The regular and gamopetalous (united) corolla, on the other hand, may be tubular, bell or funnel-shaped. The irregular flowers with polypetalous condition may have papilionaceous or butterfly-shaped corolla as in pea, whereas in gamopetalous situation the shape may be bilabiate, personate or ligulate (Fig. 16.23). In some plants, the flowers may have an appendage or an outgrowth in petals. This is called a **spur**. The mode of arrangement of petals in floral bud with respect to other members of the same whorl is known as **aestivation**. When there is no overlapping of margins as in *Calotropis* and custard-apple, it is called **valvate**. If one margin of an appendage overlaps that of the next one and so on as in china rose and cotton, it is **twisted**. There are **imbricate** and **vexillary** types of aestivations also.

Androecium is the male reproductive organ and comprises stamens. Each stamen consists of three parts, a filament (stalk), connective, and anther. Each anther is generally bilobed and has two chambers (pollen sacs) per lobe, making four chambers per anther. Each chamber is filled with pollen grains and opens through longitudinal or transverse or porous or valvular opening. In

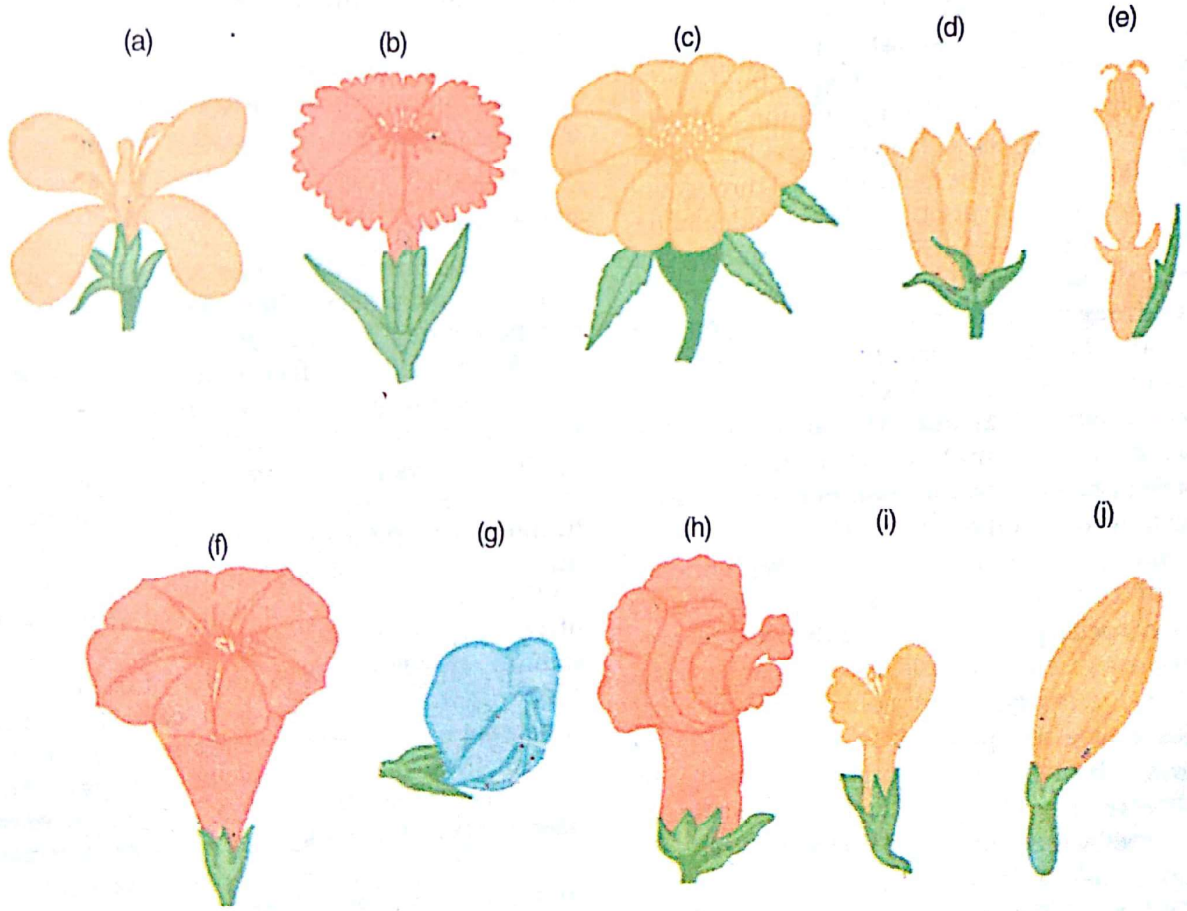

Fig. 16.23 *Types of corolla (a) cruciform (b) caryophyllaceous (c) rosaceous (d) bell-shaped (e) tubular (f) funnel-shaped (g) papilionaceous (h) bilabiate (i) personate (j) ligulate*

some flowers, *viz. Salvia* and *Verbascum*, some stamens are without pollen grains and remain sterile throughout. These are called **staminodes**. The attachment of anther to the filament (Fig. 16.24) may be **basifixed** (if attached to the base)

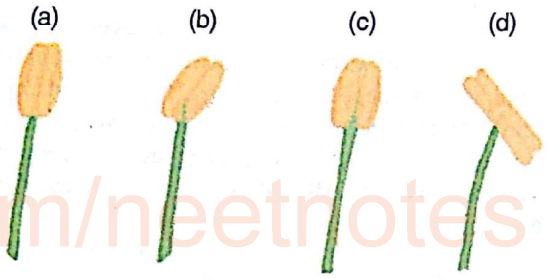

Fig. 16.24 *Attachements of anther (a) basifixed (b) dorsifixed (c) adnate (d) versatile*

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or **dorsifixed** (attached to the back of the anther making the anther immovable on the filament).

In *Michelia* and *Magnolia*, the filament runs up the entire length of the anther. This attachment is named **adnate**. It is described as **versatile** if filament is attached to the back of the anther at one point only and the anther is movable as in grasses. The stamens in a flower may either remain free (polyandrous) throughout or get united to a variable degree (adelphous, syngenesious, synandrous). Stamens are **epipetalous**, if they are attached to petals; **gynandrous** if united with carpels, or **epiphyllous** when attached to the perianth. There may be a variation in the length of stamens. In Lamiaceae, the condition is **didynamous** (four stamens, two short and two long), whereas in crucifers **Brassicaceae** it is **tetradynamous** (six stamens, inner four long and outer two short).

Gynoecium (pistil) is the female reproductive part of the flower and is made up of one (monocarpellary) or more carpels (polycarpellary). Each carpel usually consists of an enlarged basal portion, the **ovary**, which contains one or more ovules and a receptive surface for pollen, the **stigma**. The ovary and stigma are connected by a more or less elongated tube, the **style**. In polycarpellary condition, carpels may be free (apocarpous) as in lotus and rose, or united together (syncarpous) as in tomato and mustard. Each ovary bears one or more ovules attached to a flattened, cushion-like placenta (pl. placentae). The apocarpous pistils usually have one chamber (locule) containing ovules, while many syncarpous pistils have ovaries partitioned into two or more chambers. Some syncarpous pistils, like those in grasses or sedges, have only one chamber, partitions between the fused parts being absent. After fertilization, the ovules develop into seeds and the ovary matures into a fruit. A sterile pistil is known as **pistillode**. The position of style may be **terminal** (arising from the tip of the ovary), **lateral** (arising from the side of the ovary) or **gynobasic** (arising from the depressed centre of the four-lobed ovary, or directly from the thalamus). The terminal style occurs in most plants, lateral in a few, such as mango, and gynobasic in *Ocimum* and other members of the Lamiaceae. There is a variation in the shape and size of stigma. In syncarpous gynoecium, the stigma may have only one flattened disc, divided into a number of lobes. Generally, the number of lobes corresponds to the number of carpels in the gynoecium. There may be a complete cohesion at the ovary, style and stigma levels or these may be free at any one or two levels.

Placentation

The arrangement of ovules within the ovary is known as placentation. The placentation can be divided into marginal, axile, parietal, basal, central and free-central types (Fig. 16.25). In **marginal** placentation the ovules are attached along the junction of two margins of the ovary wall and there is no true placenta. In such cases, the ovary is one-chambered (unilocular), and is generally found in pea and other leguminous plants. If the ovules present on the placentae develop from the central axis of the ovary as in lemon, china rose and tomato, the ovary is multilocular, and the placentation is **axile**. In crucifers and *Argemone*, the ovules develop on the inner wall of the ovary or on peripheral part, and the placentation is **parietal**. In **free-central placentation** the ovules are borne on a central axis, septa are absent, hence the ovary is unilocular as in members of Caryophyllaceae (*Dianthus*). In **basal** placentation, the placenta develops at the base of the ovary and bears a single ovule as in the members of Asteraceae family.

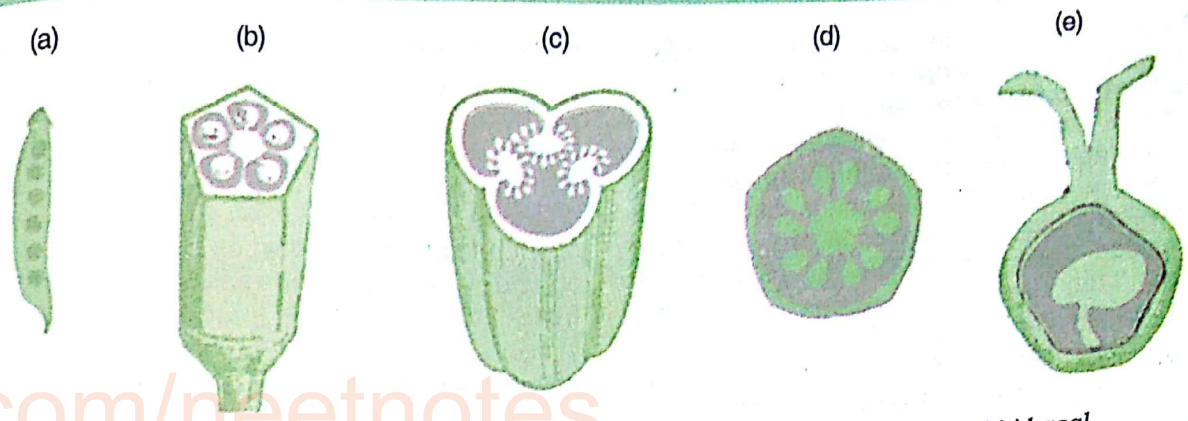

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Fig. 16.25 Types of placentation (a) marginal (b) axile (c) parietal (d) free-central (e) basal

16.6 THE FRUIT

The fruit is a mature or ripened ovary, developed after fertilization. Generally, it consists of a wall or pericarp, and seeds. The pericarp may be dry or fleshy; when fleshy it consists of outer (epicarp), middle (mesocarp) and inner (endocarp) walls. In many fruits, the division of pericarp may not be clearly visible. Sometimes, the other floral parts such as thalamus, receptacle or even calyx may develop as a part of the fruit as in apple (thalamus) and cashewnut (peduncle). These are called **false fruits**. The fruits which develop only from ovary are called **true fruits.**

Types of Fruits

There is a great variation in the shape, size and even structure of fruits. Broadly, they are divided into three groups on the basis of their origin and development. The fruits developed from the single ovary are said to be of **simple** type. If there is a cluster of simple fruits which is borne on the same thalamus and developed from polycarpellary, apocarpous ovary, they are **aggregate** as in *Calotropis*, larkspur and custard-apple. When the fruit develops from the inflorescence, it is **composite** as in pine-apple and mulberry (Fig. 16.26).

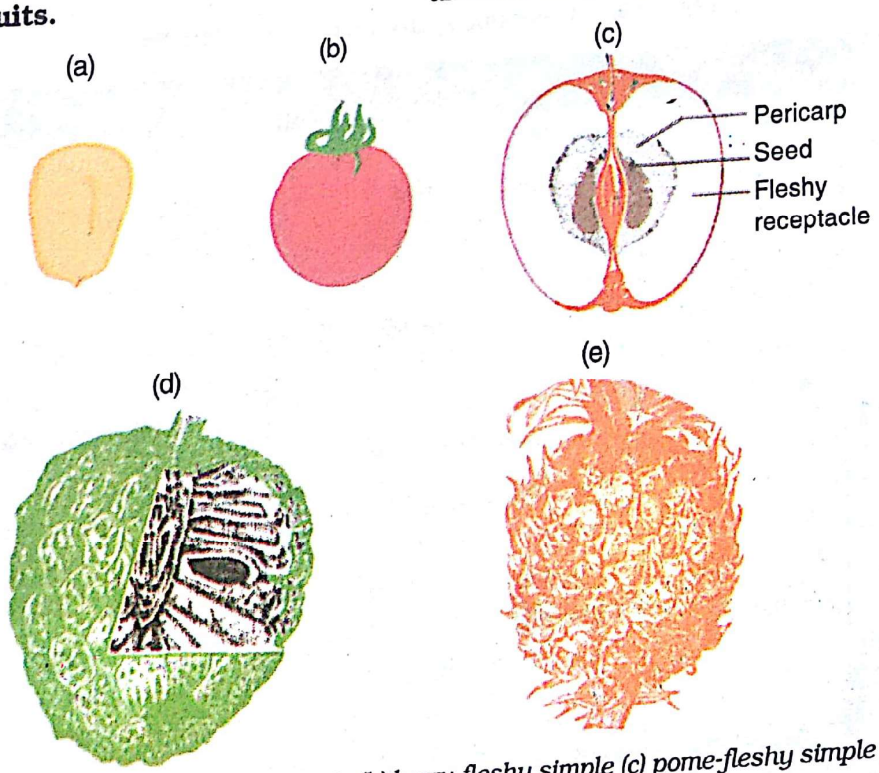

Fig. 16.26 Types of fruit (a) dry simple (b) berry-fleshy simple (c) pome-fleshy simple (d) etaerio of berries – aggregate (e) sorosis- composite

In pea, maize, wheat, marigold, *Acacia* and coriander, the fruit wall or pericarp is dry and not differentiated into epicarp, mesocarp and endocarp. such fruits are called dry fruits. On the other hand, in mango, tomato, apple, orange and pomegranate, the fruit wall is differentiated into epicarp, mesocarp and endocarp; such fruits are called **fleshy** fruit. The dry fruits which dehisce after ripening are grouped into legume (pea), follicle (*Calotropis*), siliqua (mustard) and capsule (cotton) types. The indehiscent or achenial fruits are of caryopsis (wheat), achene (*Boerhaavia*), cypsela (marigold), samara (*Shorea*) and nut (oak) types. In some cases, the indehiscent fruits split into many one-seeded parts after complete ripening. These are called schizocarpic fruits as in *Coriander* and *Ocimum*.

The fleshy fruits may be one or more chambered with one or more seeds and may develop from mono- or bi- or polycarpellary, syncarpous ovary. The fruit wall is differentiated into epicarp (skin of the fruit), mesocarp (fleshy and often edible region) and endocarp. The fleshy fruits are classified into **drupe** (endocarp hard and stony) as in mango and plum, **berry** (endocarp pulpy) as in tomato and guava, or special types of berries like **pepo** (cucurbits), **pome** (apple and pear), **hesperidium** (orange and lemon), **balausta** (pomegranate) and **amphisarca** (bel).

The aggregate fruits develop from bi- or polycarpellary, apocarpous ovary. Each ovary develops into a fruitlet. These fruitlets are borne on the thalamus and fall off as a unit. A cluster of such fruitlets is known as etaerio. It may be etaerio of follicles (*Calotropis*), etaerio of achenes (rose), etaerio of drupes (raspberry) and etaerio of berries (custard-apple) types. The composite or multiple fruits may be of **sorosis** type as in pine-apple, screwpine, jack-fruit and mulberry, or **syconus** type as in banyan, fig and peepal. Table 16.1 lists some common fruits, their type and edible part.

Dehiscence of Fruits

The dehiscent fruits rupture to liberate their seeds in several ways. In *Portulaca* and *Celosia*, the seeds are liberated through transverse rupture; in poppy and *Luffa* through pores, in pea and bean through sutural valves, in cotton and lady's finger through locules, in linseed and mustard through septa or partition wall. In plants like datura, the fruits rupture loculicidally throwing valves away from fruit, and leaving seeds attached to the central axis.

Table 16.1 Common Fruits and their Edible Parts

S.No	Name	Type	Edible part
1.	Apple	Pome-fleshy simple	Fleshy thalamus
2.	Banana	Berry-fleshy simple	Mesocarp and endocarp
3.	Cashew nut	Nut-indehiscent simple	Peduncle and cotyledons
4.	Coconut	Drupe- fleshy simple	Endosperm
5.	Custard apple	Etaerio of berries-aggregate	Pericarp
6.	Date palm	Berry- fleshy simple	Pericarp
7.	Jack-fruit	Sorosis- composite	Bracts, perianth and seeds
8.	Fig	Syconus-composite	Fleshy receptacle
9.	Guava	Berry-fleshy simple	Thalamus and pericarp
10.	Mango	Drupe-fleshy simple	Mesocarp
11.	Wheat	Caryopsis-indehiscent simple	Starchy endosperm
12.	Orange	Hesperidium- fleshy simple	Juicy placental hair
13.	Tomato	Berry- fleshy simple	Pericarp and placentae
14.	Pear	Pome- fleshy simple	Fleshy thalamus
15.	Pine-apple	Sorosis- composite	Outer portion of receptacle
16.	Litchi	Nut fleshy simple	Fleshy and juicy aril

16.7 THE SEED

The seeds develop from ovules inside the ovary after fertilization. There is a great variation in the size, shape and period of viability of seeds. A seed is generally made up of seed coat and enclosed inner parts like cotyledons, plumule, and radicle (Fig. 16.27). The cotyledons store reserve food, to be used during resting period and germination. The plumule gives rise to shoot and the radicle to root system.

On the basis of the number of cotyledons, the seeds are divided into two types, *viz* **monocotyledonous** and **dicotyledonous.** The dicot seeds are further divided into endospermic and non-endospermic, based on the presence or absence of endosperm (reserve food tissue). The endospermic seeds of castor and custard-apple contain an outer hard, blackish and mottled shell called **testa** or the **seed coat.** There is an outgrowth at the micropyle called caruncle, which is spongy and absorbs moisture readily during seed germination. A ridge called raphe, formed by the funicle, is prominent. The perisperm, a remnant of the nucellus, is a thin white papery membrane surrounding the endosperm. **Endosperm,** a fleshy food storage tissue, rich in oil is present under the cover of the perisperm. Embryo lies embedded inside the endosperm, and contains two thin cotyledons hinged to an axis called tigellum. The tigellum shows a protruding radicle and the plumule hidden between the cotyledons. The non-endospermic seeds of gram, pea and bean are also covered by a seed coat. The seed coat is generally two layered. The outer one is called **testa** and the inner **tegmen**. **Hilum** is a point of attachment of seed with the stalk. A minute opening (micropyle) above the hilum and an outgrowth (raphe) are also distinct. The embryo consists of an axis (tigellum) and two fleshy **cotyledons** full of reserve food material. The lower pointed end of the axis is **radicle** and the upper leafy end is the **plumule** (Fig 16.27).

The monocotyledonous seeds are with a single cotyledon and are generally endospermic. The seed coat is membranous, and fused with the fruit wall. The main bulk of the grain is endosperm, which stores food. Endosperm is separated from the embryo by a distinct layer known as **aleurone layer**. The embryo is small and occurs in a groove at one end of the endosperm. It consists of one shield- shaped cotyledon known as **scutellum**, a short axis with plumule and radicle. The plumule is covered by a sheath called **coleoptile.** Similarly, the radicle is protected by **coleorhiza**. Some monocots are non-endospermic, e.g. orchids and *Sagittaria.*

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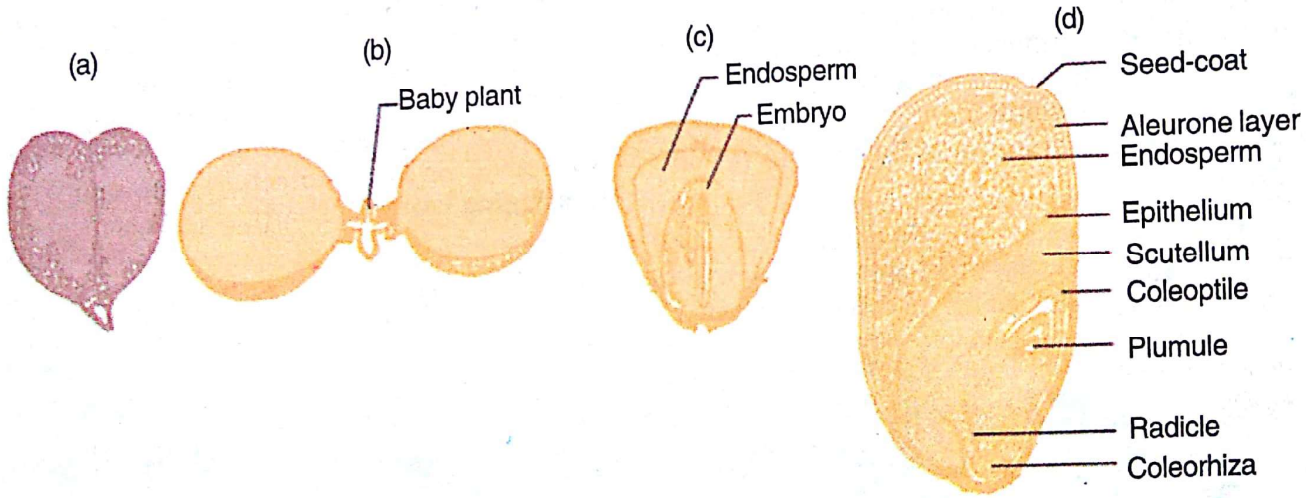

Fig. 16.27 *Structure of seeds (a, b) dicot seed (c, d) monocot seed*

16.8 DISPERSAL OF FRUITS AND SEEDS

We know that most of the plants do not move from one place to another. They grow, produce flowers and fruits while remaining fixed at one and the same place. The seeds falling directly under the mother plant have to germinate and develop under limited food supply and space. To overcome this problem, the fruits and seeds have developed several special devices for wide dispersal. The natural agents like wind, water and animals, and even mechanism of dehiscence in fruits of some plants, help the seeds and fruits to disperse from one place to another, and to long distances from the parent plant (Fig. 16.28).

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Wind

In the species where the seeds are light in weight or have some accessory part to help dissemination, are dispersed by the air current. The seeds of drum-stick and cinchona, and fruits of yam, maple and sal tree, are provided with one or more appendages in the form of thin, flat and membranous wings, which help them to float in the air and be carried away to long distances. In the members of Asteraceae, the calyx is modified into hair-like structures called pappus. They persist in fruit and open out like umbrella, helping the seeds to float in the air. In poppy and prickly poppy (*Argemone*), the fruit dehisces and seeds are thrown out to a distance away from the parent plant. The seeds of *Calotropis*, *Alstonia* and cotton are provided with hair and cover sufficient distances alongwith the wind. The seeds of orchids and some grasses are very small and light in weight and may be easily carried away by wind to far off places.

Water

The fruits and seeds with specialised devices which may be in the form of spongy and fibrous outer walls as in coconut and double coconut, spongy thalamus as in lotus, and small seeds with airy aril as in water lily, float very easily in water, and are carried away to long distances with the water current. The double coconut (*Lodoicea maldivica*) is a native of Seychelles islands and its fruits reach even the coastal regions of India.

Animals

The fruits and seeds with hooks, spines, bristles, stiff hair, etc., get attached to the body of hairy and woolly animals and are carried away by them to distant places. For instance, fruits of *Xanthium* and *Urena* bear curved hooks, spear grass has a bunch of stiff hair, *Tribulus* has sharp and rigid spines, *Boerhaavia* has sticky hair, which help their dispersal by animals. The edible fruits like guava, grape, fig and plum are dispersed by birds and even human beings, either by feeding on them and passing out undigested seeds with faeces or by carrying them to other places for later feeding.

16.9 DEFENSE MECHANISMS IN PLANTS

Plants have developed special organs or devices to repulse or avoid the attack of their enemies. Some plants like lemon, pomegranate and *Duranta* have **thorns**; pineapple, datepalm, *Agave* and *Yucca* have sharp-pointed **spines** at the leaf ends; silk cotton tree and rose have **prickles**; *Opuntia* and other cacti have spines for their protection from animals. The **stinging hair** with sharp and siliceous apex

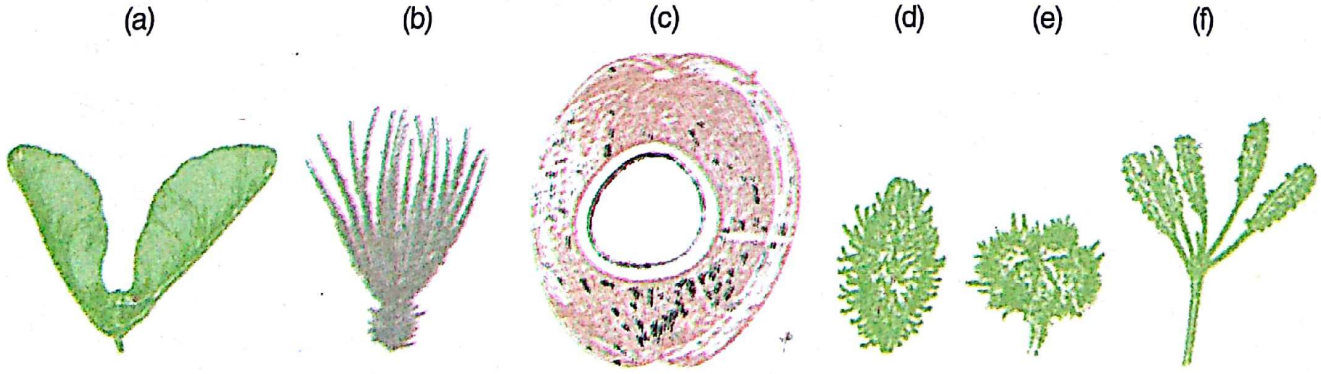

Fig. 16.28 *Dispersal of fruit and seed (a, b) by wind (c) by water (d-f) by animals*

occur on all parts of the body in nettles (*Laportea* spp) and *Urtica dioica*. **Glandular hair** with sticky substances are present in *Jatropha*, *Boerhaavia* and tobacco. The dense coating of hair or **stiff hair** which are always repulsive to animals are found in cud-weed (*Gnaphalium*) and in many cucurbits.

There are other defense devices like the presence of poisonous and irritating substances in the plants. These are in the form of latex in *Ficus*, *Nerium* and *Euphorbia*; alkaloids in poppy, *Datura* and tobacco, and irritating substances in *Colocasia* and other aroids. The plants of neem and karela have a bitter taste. Production of tannin, resin, essential oils, etc., in some plants and the **geophilous** habit in others (e.g., ginger, turmeric, colocasia and onion) are protective measures. Some plants like guava, mango and litchi have a habit of harbouring ants (**mermicophily**), which save the plant from damage by other animals. **Mimicry** is a habit of imitating the general appearance, colour, shape of other plants or animals, generally disliked by attackers. The aroids (*Caladium*) and *Sansevieria* resemble spotted snakes and are thus able to scare away plant-eating animals.

16.10 DESCRIPTION OF A TYPICAL ANGIOSPERMOUS PLANT

Various morphological features mentioned earlier, should effectively help to describe a flowering plant. The description has to be brief, in a simple and scientific language, and presented in a definite sequence. The description should help in identifying and assigning a plant to its appropriate taxonomic position.

The plant is described beginning with its **habit**, such as herb, shrub or tree, and **life-span**, annual, biennial or perennial. The **habitat** is its natural abode, and a species may be mesophyte, hydrophyte or xerophyte. The **roots** are described on the basis of their position, like primary or secondary, aerial or terrestrial; and morphology, like tap or fibrous. The **stem** is described in respect to texture (herbaceous or woody), modifications (tendril, prickle, etc.), surface (smooth, hairy, spiny, prickly, etc.); shape (cylindrical, angular or flattened). **Leaves** are described for duration (deciduous or persistent); phyllotaxy (alternate, opposite or whorled); parts (petiolate or sessile, stipulate or exstipulate, ligulate, etc.), shape (linear, lanceolate, oblong, reniform, cylindrical, etc.), margin (entire, spiny, serrate, lobed, etc.), apex (acute, obtuse, etc.), venation (reticulate or parallel, unicostate or multicostate), surface (smooth, rough, hairy, etc.), blade (simple or compound, pinnate or palmate, etc.). The description of **inflorescence** includes its type (cymose/racemose) and sub types.

The **flower** may be sessile or pedicellate, bracteate or ebracteate, unisexual or hermaphrodite, if unisexual staminate or pistillate; zygomorphic or actinomorphic, hypogynous, perigynous or epigynous, complete or incomplete, isomerous or heteromerous, if isomerous tri- tetra- or pentamerous. **Calyx** is described in terms of number of sepals, polysepalous or gamosepalous, colour, shape, aestivation and their duration on the thalamus (deciduous or persistent). The **corolla** is described with respect to the number of petals, free or united, colour and shape, aestivation, and special appendages. **Androecium** is described by taking into consideration the number of stamens, polyandrous or united, if united adelphous, syngenesious or synandrous; length of filament, anther and its shape and attachment. This is followed by the details pertaining to the female reproductive part or **gynoecium**. It includes information with regard to the number of carpels, free or united, position of ovary (superior or inferior), placentation and number of ovules per locule, stigma and style, etc. The fruits and seeds are also described by their types, origin and development.

Having described various parts of a plant, a diagram of various floral parts and a formula for these floral parts are presented. A **floral diagram** provides information about the number of parts of a flower, their general structure, arrangement, and the relation they have with one another, cohesion and adhesion, position of the flower with respect to the mother axis. It is a ground plan of a flower. The calyx lies outermost, corolla next to calyx, androecium next to corolla and gynoecium in the centre. The cohesion and adhesion in the parts, such as sepals, petals, stamens and carpels of different whorls including placentation, are also shown in the diagram.

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A dot on the top of the floral diagram represents the position of the mother axis. The floral characters of an individual plant (*Cassia fistula* - amaltas) can be represented by the following floral diagram.

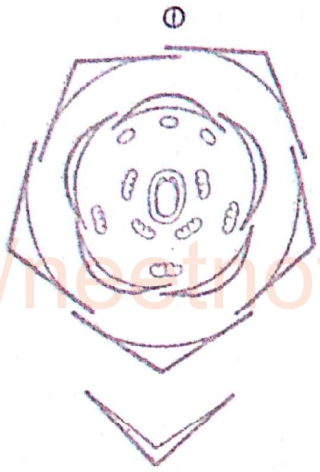

Fig. 16.29 Floral diagram of Cassia fistula (Amaltas)

The **floral formula** represents the organisation of different whorls of the flower, their number, cohesion and adhesion, and mutual relationship if any. In the formula, the letter **K** stands for calyx, **C** for corolla, **P** for perianth, **A** for androecium and **G** for gynoecium. The figures with these symbols indicate the number, cohesion is indicated by enclosing the figure within bracket and adhesion by a line drawn above. If the ovary is superior the line is drawn below $\underline{G}$ and if it happens to be inferior the line is drawn above $\overline{G}$. In the formula, a series of symbols are also used to represent certain specific features of the flower, like ♂ for male, ♀ for female, ⚥ for bisexual plants, ⊕ for actinomorphic and ◌|◌ for zygomorphic nature of the flower. The floral formula of one species can easily represent the entire family. For example,

$\oplus ♂ K_{2+2} C_4 A_{2+4} \underline{G}_{(2)}$ for Mustard (Brassicaceae);

Br $\oplus ♀ P_{3+3} A_{3+3} \underline{G}_{(3)}$ for *Asphodelus tenuifolius* (Liliaceae)

16.11 DESCRIPTION OF SOME IMPORTANT FAMILIES

BRASSICACEAE

The family formerly known as Cruciferae, has 375 genera and about 3200 species, including 150 from India. They are distributed all over the world, particularly in Mediterranean and temperate regions.

Vegetative Characters

Plants are generally herbs, annual or biennial; taproot swollen and modified due to stored food as in radish and turnip; stem, herbaceous, erect, cylindrical, sometimes reduced, glabrous or hairy, solid and branched; leaves, simple, alternate or subopposite, exstipulate, generally sessile, hairy, with unicostate, reticulate venation.

Floral Characters

Inflorescence: raceme, or corymb; flower: pedicellate, ebracteate, hermaphrodite, actinomorphic, rarely zygomorphic as in *Iberis*, hypogynous, complete and tetramerous; calyx: four sepals in two whorls, polysepalous, imbricate aestivation; corolla: four petals, polypetalous, arranged alternately with sepals in cruciform manner, generally clawed; androecium: stamens generally six, arranged in two whorls, outer two short and inner four long – a tetradynamous condition, polyandrous, anthers introrse, basifixed, dithecous, nectaries present at the base of stamens; gynoecium: normally bicarpellary, syncarpous, unilocular becomes bilocular due to a false septum called 'replum', ovary superior, placentation parietal with several ovules, style short and stigma simple or bifid; Fruit: siliqua; seeds: exalbuminous. Fig 16.30 illustrates vegetative and floral parts, floral diagram and floral formula of mustard plant as an example for the family.

Economic Importance

The members of the family provide food, vegetables, oil, medicines and ornamental plants. Gobhi (*Brassica oleracea*), and muli (*Raphanus sativus*) are consumed as vegetable. The oil is extracted from the seeds of sarson (*Brassica campestris*), **kalirai** and **rai** and used for cooking. The material left after the extraction of oil is highly nutritious and used as a cattle feed. The leaves and tender shoot of garden cress (*Lepidium sativum*) are used in liver complaints, asthma, cough and bleeding piles. The seeds of *Cheiranthus cheiri* (wall flower) are useful in bronchitis and fever, and flowers in treatment of paralysis and impotency. *Lobularia* is said to be

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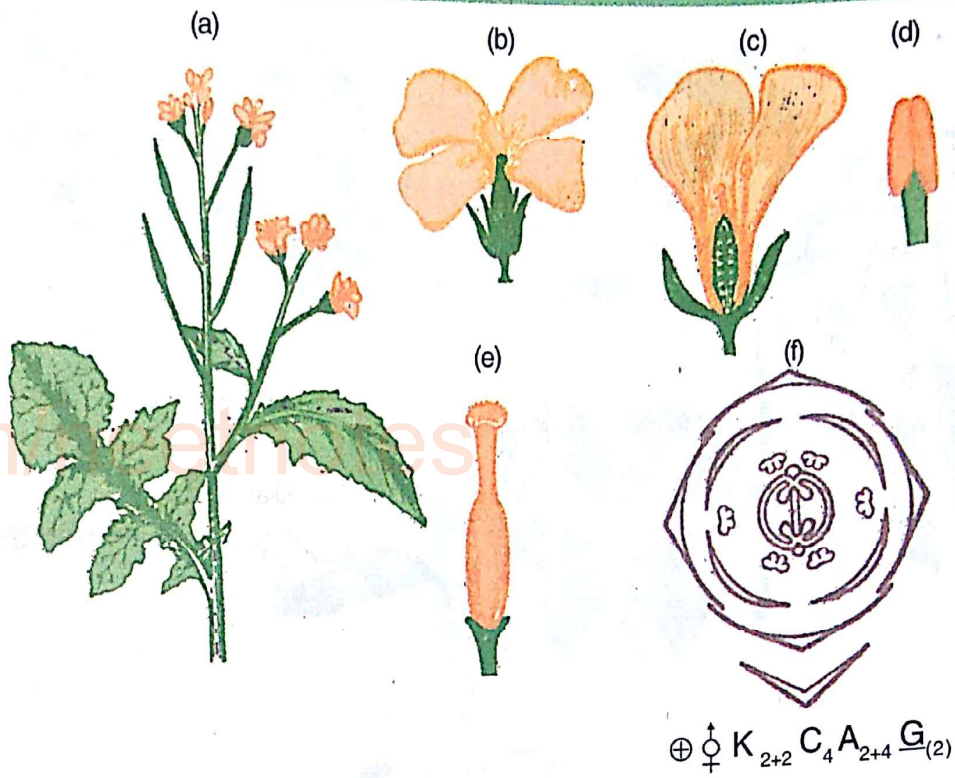

Fig. 16.30 *Brassica campestris (mustard) plant (a) flowering twig (b) a flower (c) L.S. of flower (d) stamen (e) carpel (f) floral diagram and formula*

beneficial in gonorrhoea and *Iberis amara* (chandni) in rheumatism and gout. Some plants like wall flower and chandni, are grown in gardens and parks as ornamentals for their beautiful flowers.

FABACEAE

This family was earlier known as Papilionoideae. The members of this family are of great economic value.

Vegetative Characters

Plants may be herbs, shrubs, climbers, and even trees in their habit and may occur in xerophytic, mesophytic, hydrophytic, or halophytic habitats; tap root system, much branched and with bacterial nodules; stem, erect or twin, branched and angular or cylindrical; leaves, generally compound, usually trifoliate, modified partly or wholly into tendril with pulvinate leaf base.

Floral Characters

Inflorescence: raceme, rarely solitary axillary; flowers: pedicellate, zygomorphic, hermaphrodite and complete; calyx: sepals five, gamosepalous, imbricate aestivation; corolla: petals five, polypetalous, the posterior one large and outermost, the next two lateral ones (wings) and two anterior and innermost ones united (keel or carina), aestivation descending imbricate; androecium: stamens ten, diadelphous (9+1), introrse, basifixed and dithecous; gynoecium: monocarpellary, ovary superior, placentation marginal with many ovules, style long slightly bent at the apex, flattened, stigma simple or capitate; fruit: legume, indehiscent; seed: exalbuminous with large embryo. Pollination is by insects/bees. Fig.16.31 illustrates pea as an example of the family.

Economic Importance

The family provides various types of pulses, medicines, fibres, timbers, dyes and plants for gardens. The fruits and seeds of pea, gram, arhar (*Cajanus cajan*), sem (*Dolichos lablab*), moong (*Phaseolus radiatus*), soyabean (*Glycine max*) are rich in protein, and are used as vegetables and pulses. Groundnut (*Arachis hypogea*) produces oil for us and oil cake for cattle. Muliathi (*Glycyrrhiza glabra*) is used as medicine in throat pain and cough. The fresh juice of ratti (*Abrus precatorius*) leaves is said to be useful in leucoderma, the juice of 'agast' (*Sesbania*

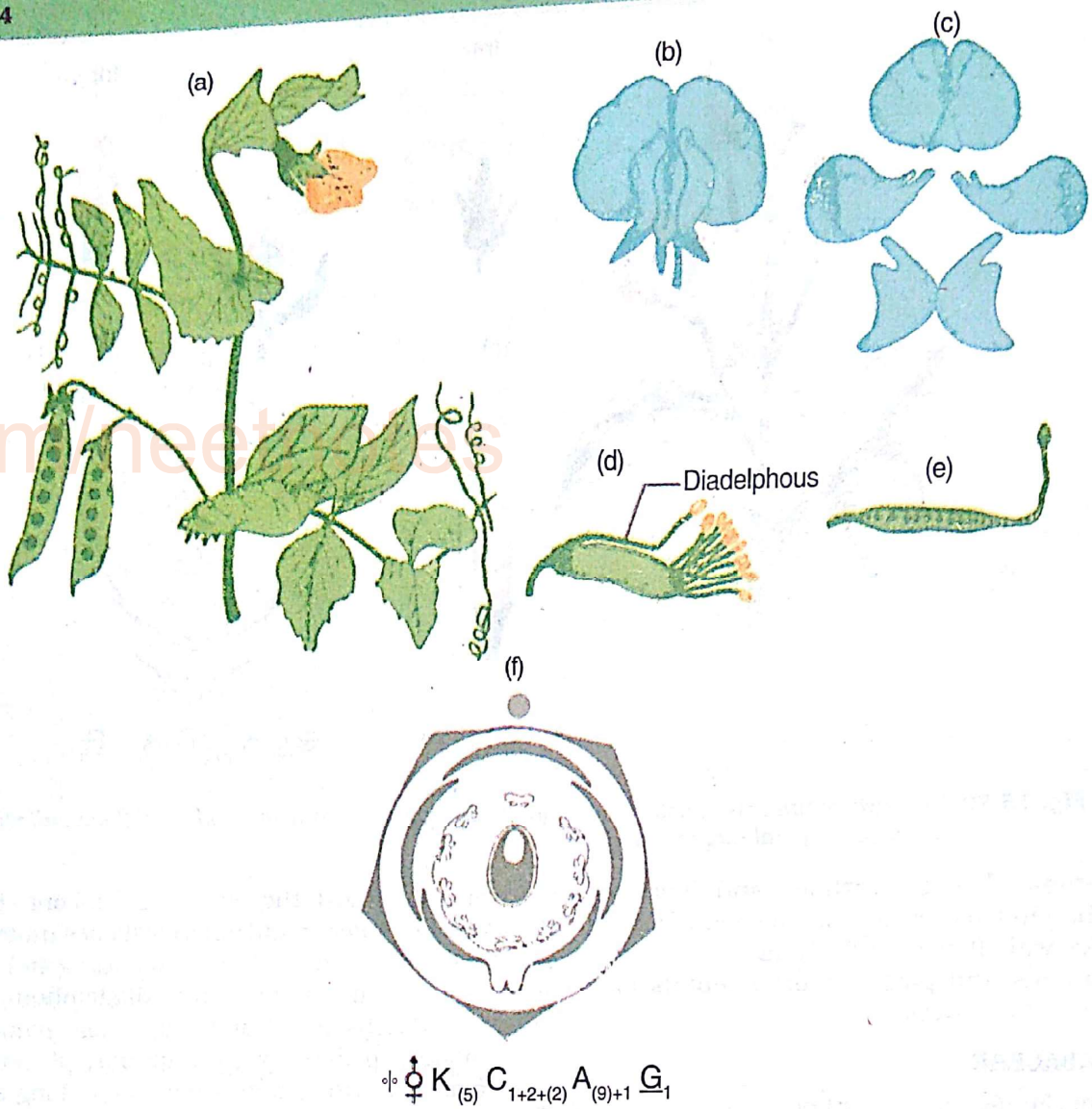

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Fig.16.31 *Pisum sativum (pea) plant (a) flowering twig (b) a flower (c) petals (d) androecium (e) L.S. of carpel (f) floral diagram and formula*

grandiflora) flowers is believed to be beneficial in improving eye sight. Sunn- hemp (*Crotalaria juncea*) yields fibres, which are used for making rope, mat, canvas, nets, sacks, etc. Shisham (*Dalbergia sissoo*) and Indian rose-wood (*D. latifolia*) yield a dye. The plants of *Lathyrus, Clitoria, Sesbania,* and *Erythrina* (Indian coral tree) are grown for ornamental purposes. Some plants like *Butea monosperma* and *Astragalus gummifer* also produce medicinally useful gum.

ASTERACEAE

It is the largest family of dicot plants comprising 950 genera and about 20,000 species including about 1000 from India. It is commonly known as the 'sunflower family' and is distributed throughout the world.

Vegetative Characters

Plants are mostly herbs or shrubs but rarely trees, occupy mesophytic, xerophytic or hydrophytic habitats. Tap root, sometimes modified into tubers (*Dahlia*); stem, erect or prostrate, generally herbaceous, rarely woody, hairy, cylindrical, glabrous, solid or fistular; leaves, simple, petiolate or sessile, arranged alternately, rarely opposite or whorled, exstipulate, unicostate or multicostate, reticulate venation.

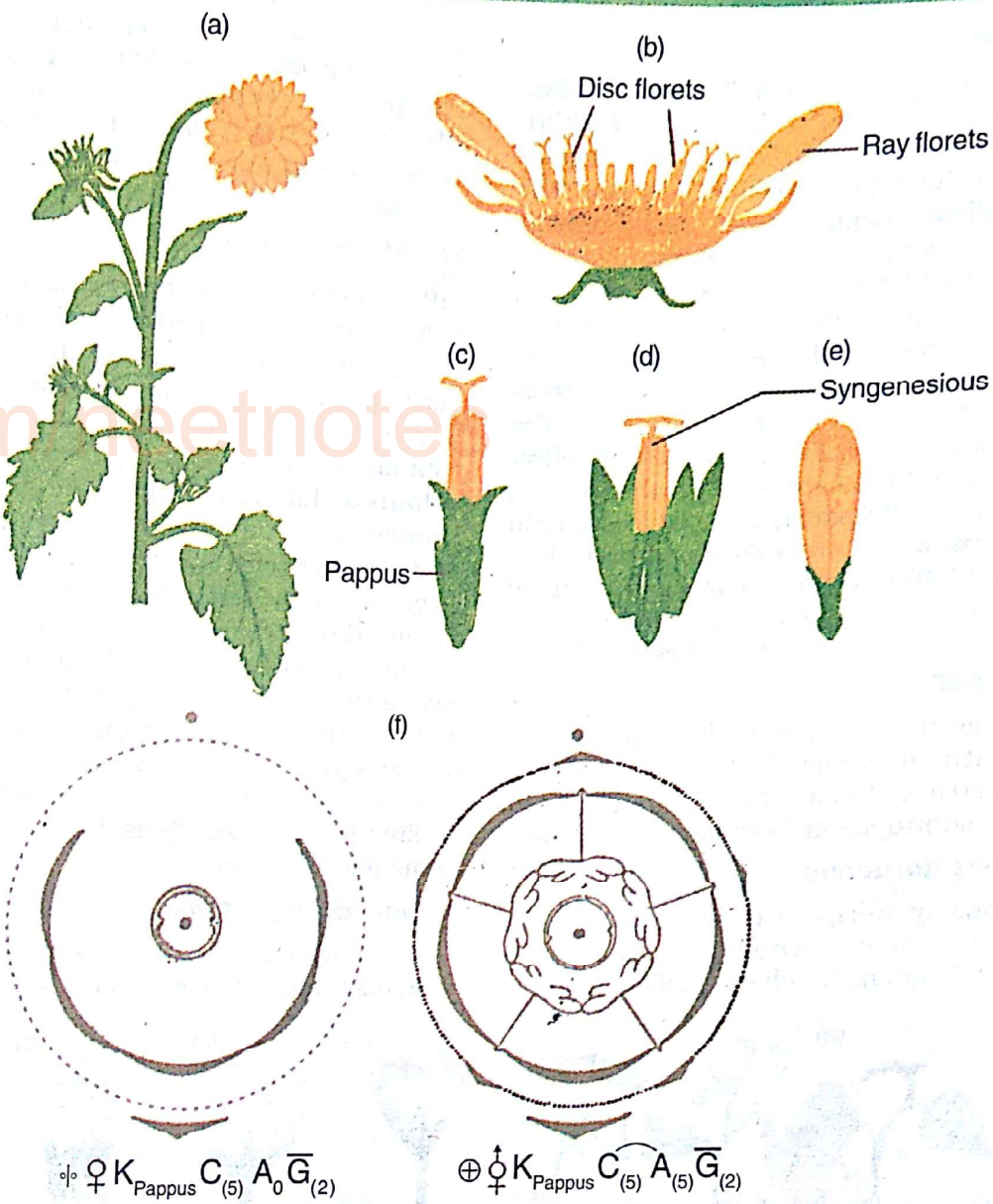

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Fig.16.32 *Helianthus annuus (sunflower) plant, (a) flowering twig (b) L.S. of inflorescence (c, d) disc floret (e) ray floret (f) floral diagrams and formulae*

Floral characters

Inflorescence: head or capitulum with ray and disc florets surrounded by involucral bracts; flower: sessile, bracteate, complete or incomplete, hermaphrodite or unisexual, pentamerous, tubular or ligulate, epigynous, actinomorphic or zygomorphic; (a) Ray Florets - zygomorphic, ligulate, neutral or pistillate, epigynous; calyx: sepals absent or modified into pappus; corolla: petals five, fused, coloured, ligulate, valvate; androecium: stamens generally absent; gynoecium: bicarpellary, syncarpous, ovary inferior, placentation basal, style one and stigma bifid; fruit: cypsela, seeds: non-endospermic. (b) Disc Florets - sessile, bracteate, complete, hermaphrodite, actinomorphic, pentamerous, epigynous and tubular; calyx: sepals modified into pappus and persistent; corolla: petals five, fused, tubular and coloured; androecium: stamens five, epipetalous, introrse, syngenesious, dithecous; gynoecium: bicarpellary, syncarpous, ovary inferior, placentation basal, style simple, long and stigma bifid; fruit: cypsela, seeds: non-endospermic. Sunflower (*Helianthus annuus*) plant is being presented as an example of the family in Fig. 16.32.

Economic Importance

The plants of the family are well-known for their ornamental value. *Zinnia*, *Dahlia*, *Chrysanthemum*, *Aster*, *Helianthus* and *Tagetes* are some of the plants grown in gardens. The seeds of safflower, sunflower and *Artemisia* yields oil used for cooking and soap-making. The roots of chicory (*Cichorium intybus*) and *Helianthus tuberosum* and leaves of *Lactuca sativa* are edible. *Artemisia* yields santonin used as vermifuge. *Solidago* is used in dropsy, the roots of *Taraxacum* are useful in bowel disorders. The juice of *Emilia sonchifolia* leaves has cooling effect and is used in eye inflammation and for curing night blindness. *Eclipta alba* is used as a tonic in spleen enlargement. *Centipeda orbicularis* is used in cold and toothache. The capitulum of *Chrysanthemum roseum* and *C. cinerariaefolium* are dried, powdered and used as an insecticide.

SOLANACEAE

It is a large family, commonly called 'potato family' with 90 genera, and 2000 species including 60 from India. It is widely distributed in tropics, subtropics and even temperate zones.

Vegetative Characters

Plants mostly herbs, rarely shrubs; stem herbaceous rarely woody, aerial, erect, cylindrical, branched, solid or hollow, hairy or glabrous, underground stem in potato (*Solanum tuberosum*); leaves, cauline or ramal, simple, exstipulate, petiolate or sessile, arranged alternately, rarely opposite, pinnatisect in tomato (*Lycopersicum esculentum*), unicostate reticulate venation.

Floral Characters

Inflorescence: solitary axillary, umbellate or helicoid cyme as in *Solanum*; flower: bracteate or ebracteate, pedicellate, complete, hermaphrodite, pentamerous, actinomorphic and hypogynous; calyx: sepals five, united, tubular or campanulate, persistent, green or coloured, hairy; corolla: petals five, fused, tubular or infundibuliform, aestivation valvate or imbricate, coloured; androecium: stamens five, epipetalous, anther introrse, dithecous, basifixed or dorsifixed, filament deeply inserted in corolla tube; gynoecium: bicarpellary, syncarpous, ovary superior, placed obliquely, placentation axile, with many ovules in each locule, style simple, stigma bifid or capitate; fruit: capsule or berry; seeds: endospermic and pollination entomophilous. Fig. 16.33 illustrates a member of the family.

Economic Importance

The family provides food, medicine including narcotics, and ornamental plants. Potato, brinjal,

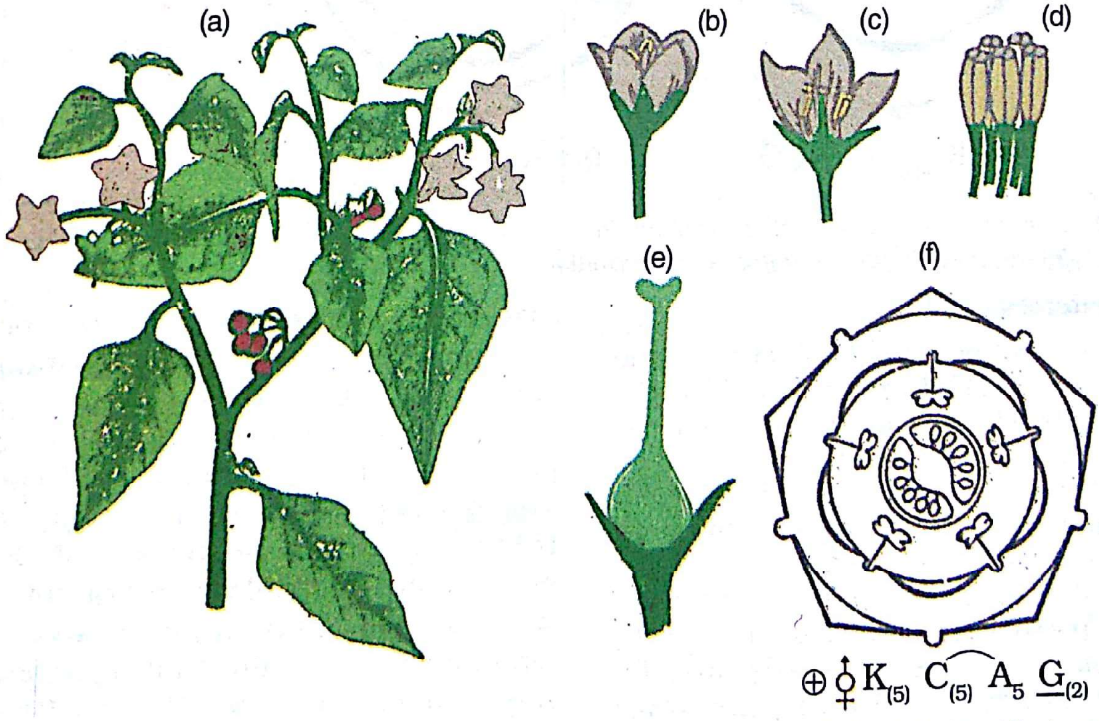

Fig.16.33 *Solanum nigrum (makoi) plant (a) flowering twig (b) flower (c) L.S. of flower (d) stamens (e) carpel (f) floral diagram and formula*

tomato, chillies are used as vegetables. Raspberry produces edible fruits. Belladona (*Atropa belladonna*) contains atropine alkaloid and used in eye testing and plaster. Nicotine obtained from tobacco is used as an insecticide. The seeds of *Datura*, Henbane (*Hyoscyamus niger*), Kateli (*Solanum xanthocarpum*) and roots of ashwagandh (*Withania somnifera*) are used medicinally. Tobacco used in bidi, cigarettes, and for chewing is obtained from the leaves of tobacco plant. Some plants like *Cestrum nocturnum* (rat-ki-rani), *Petunia*, *Schizanthus*, are grown in gardens for their beautiful flowers.

LILIACEAE

Commonly called the 'lily family' is a characteristic representative of monocotyledonous plants. It includes about 250 genera, and 4000 species, distributed worldwide. About 200 species are available in India.

Vegetative Characters

Plants mostly herbs with perennating rhizome or bulb, a few climbers (*Asparagus* and *Smilax*), *Yucca* and *Aloe* are xerophytic; root, fibrous, tuberous in *Asparagus*; stem, solid or fistular, underground rhizome, bulb or corm, aerial-climbing or erect and may have phylloclades; leaves, radical or cauline, exstipulate, alternate, opposite or whorled, sessile or petiolate with a sheathing base, venation parallel, reticulate in *Smilax*. In *Asparagus*, the leaves are reduced to minute scales (cladodes).

Floral Characters

Inflorescence: solitary axillary, panicled raceme or cymose umbel; flower: pedicellate, actinomorphic or zygomorphic, hermaphrodite or unisexual in *Smilax* and *Ruscus*, hypogynous, complete, rarely incomplete, trimerous rarely bi- or tetramerous; perianth: six in two whorls, scarious or membranous, polyphyllous or gamophyllous, petaloid or sepaloid, valvate aestivation; androecium: stamens six arranged in two whorls, polyandrous, may be epiphyllous and opposite to perianth lobes, filament long, anther dithecous, introrse or extrorse, versatile or basifixed; gynoecium: tricarpellary, syncarpous, ovary superior, placentation axile, style simple, stigma trilobed; fruit: berry or capsule; seed: endospermic. Fig. 16.34 illustrates onion plant as an example of the family.

Economic Importance

The plants like onion, garlic and *Asparagus* are used as food. *Smilax*, *Aloe*, *Gloriosa*, *Colchicum* (corms produce the dye colchicine) and *Scilla* yields useful drugs. Aloin, a purgative is obtained from *Aloe vera*, rat poison from *Urginea* and *Scilla*, tonic from *Asparagus* (shatavar). *Yucca gloriosa*

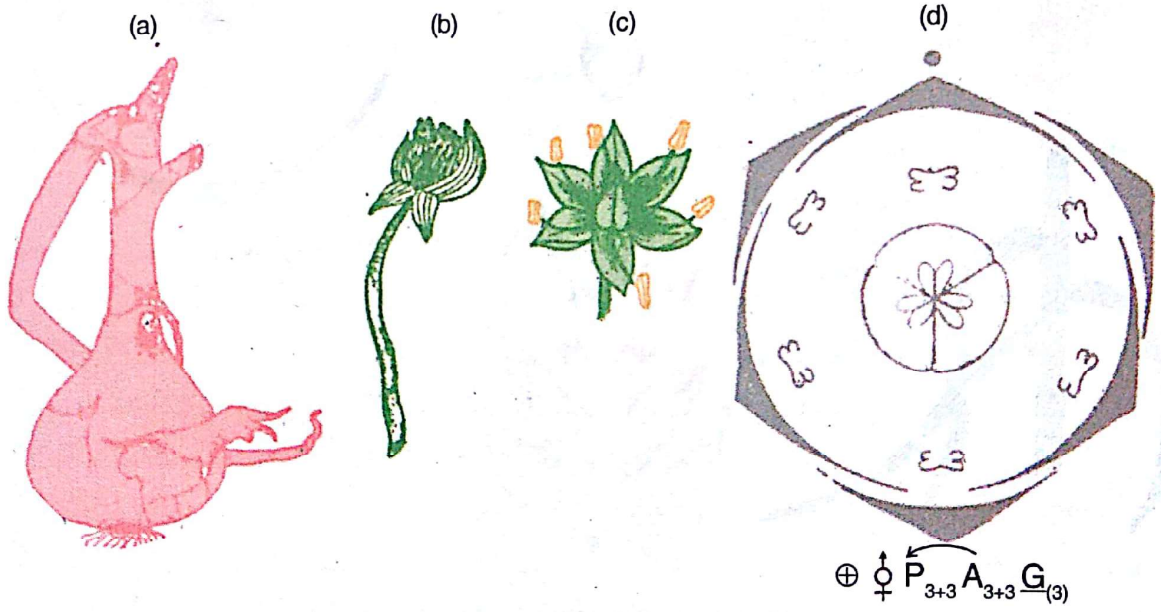

Fig.16.34 *Allium cepa (onion) plant (a) complete bulb (b) inflorescence (c) a flower (d) floral diagram and formula*

and *Phormium tenax* produce fibres, *Dracaena* and *Xanthorrhoea* yield resin, used for preparing sealing wax. The plants of *Lilium*, *Gloriosa*, *Ruscus*, *Asparagus*, etc. are cultivated in gardens.

POACEAE

The earlier name of the family was Graminae. It is economically the most important family of monocotyledonous plants. It is also one of the largest families comprising about 6000 species and 620 genera, of which about 900 species are present in India.

Vegetative Characters

Plants are generally annual herbs, although some may be perennials and shrubs, rarely trees such as bamboo; root, fibrous and branched, nearly fascicled or stilt as in maize; stem, cylindrical, with clear nodes and hollow internodes, underground rhizome as in perennial grasses; leaves, simple, arranged alternately, exstipulate, sessile, ligulate with a tubular, open sheath, developed from leaf base, venation parallel.

Floral Characters

Inflorescence: compound spike with small spikelets arranged on the main axis or rachilla, two sterile scales called glumes occur at the base of the main axis; a series of florets are present above the glume with superior and inferior palea; a long, stiff hair called awn is generally present on each lemma; flowers: bracteate and bracteolate, sessile, incomplete, hermaphrodite or unisexual, irregular and hypogynous; perianth: membranous scales called lodicules; androecium: stamens three (rarely six), polyandrous, filament long, anther versatile,

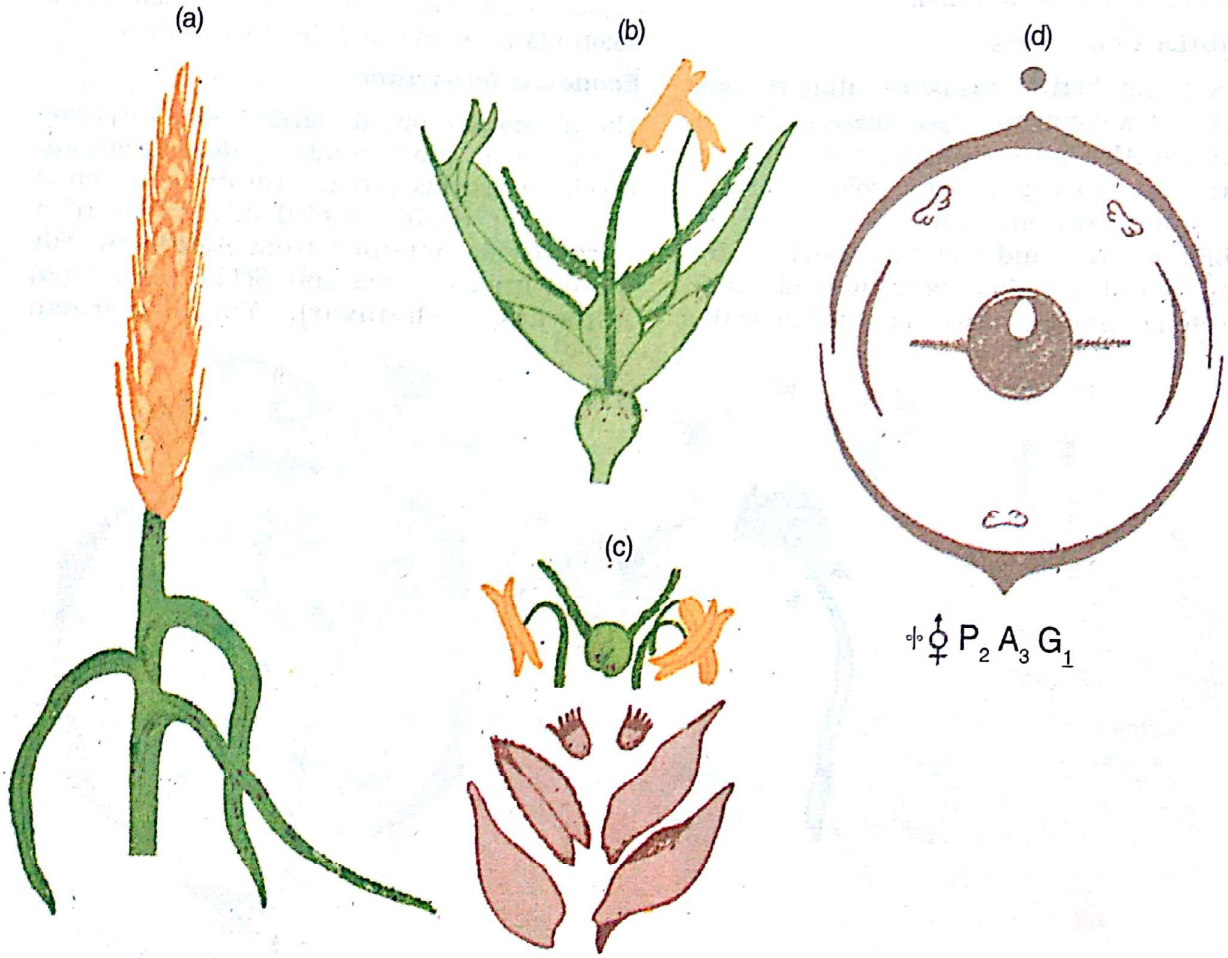

Fig.16.35 *Triticum aestivum (wheat) plant (a) flowering twig (b) a flower (c) parts of flower (d) floral diagram and formula. Note that the three carpels are united at a very early stage, giving rise to only one chamber with a single ovule.*

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long, anther versatile, dithecous, linear and extrorse; gynoecium: carpels three, syncarpous (generally carpels are fused at a very early stage forming only one chamber) with a single ovule, ovary superior, placentation basal, style short, stigma feathery or papillate and branched; fruit: caryopsis, rarely a nut (*Dendrocalamus*) or berry (*Bambusa*); seeds: endospermic with a single cotyledon. Fig. 16.35 illustrates wheat plant as a common example of the family.

Economic Importance

The staple food grains (wheat, rice, jau, jowar, oat and bajra) for majority of the human population are obtained from this family. Fodder is provided by many grasses. gur and sugar from sugarcane; and building and furniture materials from bamboos are some of the other examples of household goods from this family. We get medicine from *Cymbopogon flexuosus*. Several grasses produce volatile scented oil used in perfumery, e.g, khuskhus (*Vetiveria zizanoides*) and lemon grass. Some grasses including bamboo are used to produce paper whereas some plants are ornamental.

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SUMMARY

The flowering plants dominate the flora of Earth. There is an enormous variation in size, life-span, habit and habitat, and even mode of nutrition. The root and shoot systems are well developed and vary in shape, size, structure, and even functions. A tap root system is generally found in dicot plants and a fibrous root system in monocot plants. The roots in some plants are modified into storage organs, and for mechanical support, etc.

The presence of nodes and internodes, exogenously developed multicellular hair and positive phototropic nature, are the morphological characters that differentiate stem from root. There is a lot of variation in the shape, size and structure of the stem. Some may be strong and remain upright, as in trees and shrubs, whereas some remain weak and require support to climb on neighbouring plants or other objects. In several species, stem is modified as rhizome, corm, tuber, bulb, runner, sucker, tendril, thorn, prickle, phylloclade, bulbil, etc. to perform diverse functions under different conditions. Also stem exhibits a variety of branching pattern.

Leaf is a lateral outgrowth of the stem or branch developed exogenously at the node. The leaf-base, petiole and lamina are the parts of the leaf and exhibit marked variations in their shape and size. Even there is a variation in the features of margin, apex, surface and extent of incision of leaf blade. The venation of leaf may give an idea about the group to which the plant belongs, viz. dicot (reticulate) and monocot (parallel). The leaf may have a single leaf blade (simple type) with one midrib or may be segmented (compound type) with many leaflets. The leaves are arranged at the nodes in an organised pattern (phyllotaxy). Like other parts, leaves are also modified into different structures. There are more than one type of leaves (heterophylly) in some plants.

The flower is a modified shoot, meant for sexual reproduction. These may occur singly or in groups on a branch with limited or unlimited growth. There are different forms of inflorescence like panicle, spike, catkin, spadix, umbel, or even cyathium, verticillaster or hypanthodium. Generally, a flower has four parts namely calyx, corolla, androecium and gynoecium. There is a variation in the structure of flower and its parts, symmetry, position of ovary in relation to other parts, etc. The position of ovules may be marginal, axile, basal, parietal or central on the placenta of ovary.

The fruits are ripened ovary, made up of three layered wall which may be dry or fleshy. These may develop from a single ovary or in cluster from apocarpous ovary or even from entire inflorescence. The dry fruits are of legume, follicle, siliqua, capsule types and may dehisce after ripening, or caryopsis, achene, cypsela, samara or samaroid, and nut types which may not dehisce even after ripening. The fleshy fruits may be of drupe, berry, pepo, pome, hesperidium or even balausta types, whereas aggregate and composite fruits are of etaerios of follicles, achenes, drupes and berries, sorosis and syconus. The dehiscent fruits are ruptured to liberate seeds through transverse, pore, sutural valve, loculus or even partition wall.

Seeds are developed from ovules after fertilisation and vary in shape, size and period of viability. They are made up of cotyledons, plumule and radicle, enclosed by a seed coat. Most of the dicotyledon seeds do not have endosperm, whereas it is present in monocotyledon seeds. In endospermic seeds the plumule and radicle are protected by coleoptile and coleorhiza, respectively. Some fruits and seeds develop special devices for their dispersal from one place to another; wind, water and animals including man, help in the dispersal of seeds. Plants develop structures like thorns, spines, prickles, stinging or glandular or stiff hairs and even geophilous or mimicry habits to protect themselves from exploiters.

An angospermic plant is described in a definite sequence by using scientific terms. The plant is described for its habit and habitat, various structural features of root, stem, inflorescence, flower, fruit and seed, and their modifications. The floral features are also presented in the summarised form as floral diagram and floral formula. Scientific description of a few selected families, covering plants of high economic value, has been given as sample for study.

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EXERCISES

1. Describe the various parts of an angiospermic plant with a well-labelled diagram.
2. Write short note on the following modifications of roots: Fusiform, Pneumatophore, Tuberous, Prop, sucking root.
3. "Potato is a stem and sweet potato is a root" justify the statement on the basis of external features only.
4. Write a note on the patterns of branching in stem and their significance.
5. What are the differences between a thorn and prickle? Describe with examples.
6. Phylloclade and cladode are aerial modifications of stem. Justify with examples.
7. Write short notes on phyllotaxy and heterophylly.
8. "Leaves are modified into tendril, spine, scale, phyllode, pitcher and bladder to perform special function". Illustrate the statement with suitable examples and diagrams.
9. What do you understand by special types of inflorescence? Describe them with diagrams and examples.
10. "Flower is a modified shoot" justify the statement.
11. What do you understand by staminode and pistillode?
12. Describe in brief the various types of placentations found in flowering plants.
13. What is aestivation? Describe its various types found in petals.
14. Differentiate between
    (i) stipule and bract
    (ii) Introrse and extrorse anther
    (iii) Apocarpous and syncarpous ovary
    (iv) Sepaloid and petaloid calyx
    (v) Actinomorphic and zygomorphic flower
15. Write an account on various types of fruits.
16. Describe the edible part of the following fruits:
    Mango, grape, tomato, banana, pineapple, apple.
17. What do you understand by the dispersal of fruits and seeds? Describe the role of various agents in it.
18. Give a scientific description of family Brassicaceae. Mention the economic importance.

CHAPTER 17

INTERNAL STRUCTURE OF ANGIOSPERMOUS PLANTS

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In the earlier chapter you have studied the different parts of a plant. The plant body is made up of cells, which organise themselves into different types of tissues. The different types of tissues organise themselves in tissue systems. They reveal a definite structural and functional organisation. We can understand the definite structural and functional organisation of tissue system by studying the internal structure of different parts of the plant. In this chapter we will study about tissue, tissue system, internal structure of dicot and monocot stem, root and leaf of flowering plants and also the secondary growth in them.

17.1 THE TISSUES

Plant tissues may be classified into two main groups: (i) meristematic tissues, and (ii) permanent tissues.

Meristematic Tissues (Meristems)

All the cells of the embryo of a plant are capable of division, however, with the growth of the plant this feature becomes restricted to only certain regions. A meristem (*Gk. Moistos* : divisible) is a localised region in which actual cell division occurs. According to their origin and development, meristems are classified as **promeristem, primary meristem** and **secondary meristem.** The meristem may also be classified according to their position in the plant body as **apical, intercalary and lateral** (Fig. 17.1).

Apical meristem lies at the apex of both the stem and the root. This usually exhibits a dome-shaped structure with clear demarcation in the outer layers (**tunica**) and the inner mass (**corpus**) (Fig. 17.2).

The apical meristem is composed of a small mass of cells which are alike and constitute the promeristem. The cells of the promeristem

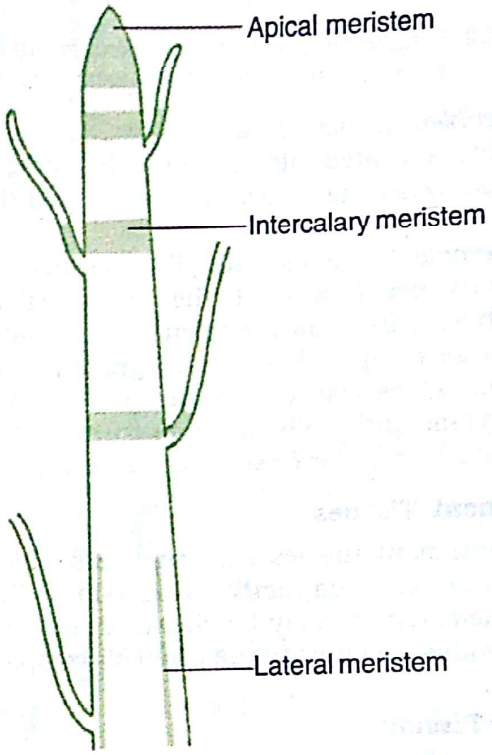

Fig. 17.1 Characterisation of meristems on the basis of location

differentiate into three regions, *viz.* **dermatogen, periblem** and **plerome**, which grow and give rise to primary permanent tissues.

(a) Dermatogen (*L. derma* : skin; *gen* : producing) : This is the single, outermost layer of cells. These divide and give rise to the skin layer (epidermis) of the stem. In roots, the cells of the dermatogen form a mass of tissue called **calyptrogen** (*L. calyptra* : cap; *gen* : producing). The calyptrogen is also meristematic and gives rise to the root cap. In dicotyledonous plants, the dermatogen forms the outermost layer (epiblema) of the root.

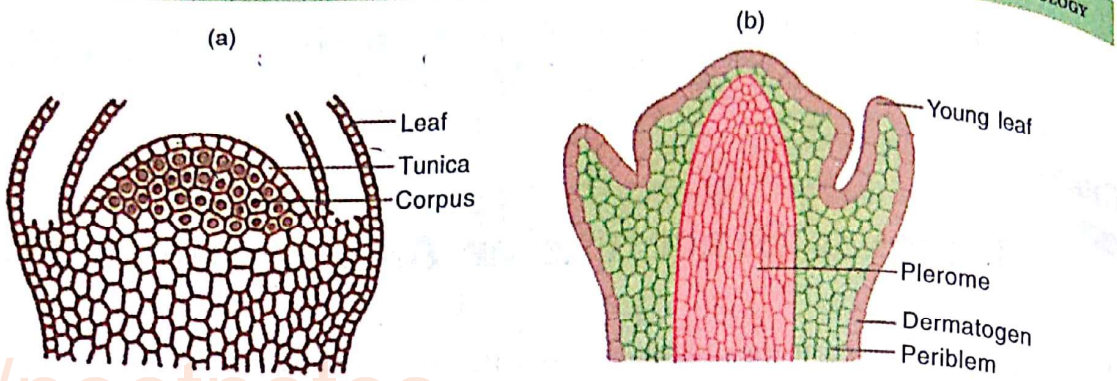

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Fig. 17.2 *Longitudinal section of a typical apical meristem (a) shows tunica layer and corpus mass (b) shows three regions – dermatogen periblem and plerome*

(b) *Periblem* (*L. peri :* around; *blema :* covering): This is located internal to the dermatogen and forms the cortex of the stem and the root.

(c) *Plerome* (*L. pleres :* full) : This lies internal to the periblem and is the central region, where cells show a tendency to elongate. These elongated cells form **procambium** that gives rise to the vascular tissues (xylem and phloem) constituting the central cylinder or stele of the stem.

Permanent Tissues

The permanent tissues comprise cells which have lost their capacity of division. The permanent tissues may be classified into two main groups (i) simple tissues, and (ii) complex tissues.

Simple Tissues

These are homogeneous in nature and are composed of structurally and functionally similar cells. The simple permanent tissues are **parenchyma**, **collenchyma** and **sclerenchyma**. Parenchyma is the most common tissue which is morphologically and physiologically unspecialised, forms the framework of all plant organs and tissues like cortex, pith, mesophyll of leaf, and floral parts. Parenchymatous cells are usually isodiametric but may be elongated or even lobed as in the mesophyll tissue of the leaves. These may either be closely packed or show small intercellular spaces (Fig. 17.3a). The **plasmodesmata** (the thread-like cytoplasmic strands, running from one cell to other) are commonly present. When the parenchymatous cells are exposed to light, these develop chloroplast in them and such a tissue is known as chlorenchyma. The cells of parenchyma are involved in the various physiological activities like photosynthesis, assimilation, storage, secretion, excretion, etc.

The collenchyma is another simple tissue composed of more or less elongated cells with primary, non-lignified cell walls. Their cell wall is the most distinctive feature which is characteristically unevenly thickened (Fig. 17.3b). The wall thickening is primary in nature and is composed of cellulose, hemicellulose and pectic materials with a high percentage of water. The thickenings may be primarily at the corners or angles of the cells. These have vacuolated protoplasts and occur characteristically in the hypodermis (the layer lies below the epidermis) of herbaceous dicots, either as a homogeneous layer or in patches. These cells constitute an effective mechanical tissue and provide elasticity and support to the growing organs.

Sclerenchyma is the third simple tissue meant specially for mechanical support. These are considerably thick-walled and lignified with simple or bordered pits in their walls and are characterised by the absence of living protoplast. On the basis of variation in form, structure, origin and development, these may be either **fibres** (Fig. 17.3c) or **sclereids** (Fig. 17.3d). The former are pointed and needle-like. The fibres occur in groups, as sheets or as cylinders in various parts of the plant body. Sclereids are very thick walled, hard and strongly lignified. They are mostly isodiametric, polyhedral, short and cylindrical. These are dead cells with very narrow cell cavities (lumen) due to excessive thickness of the cell wall.

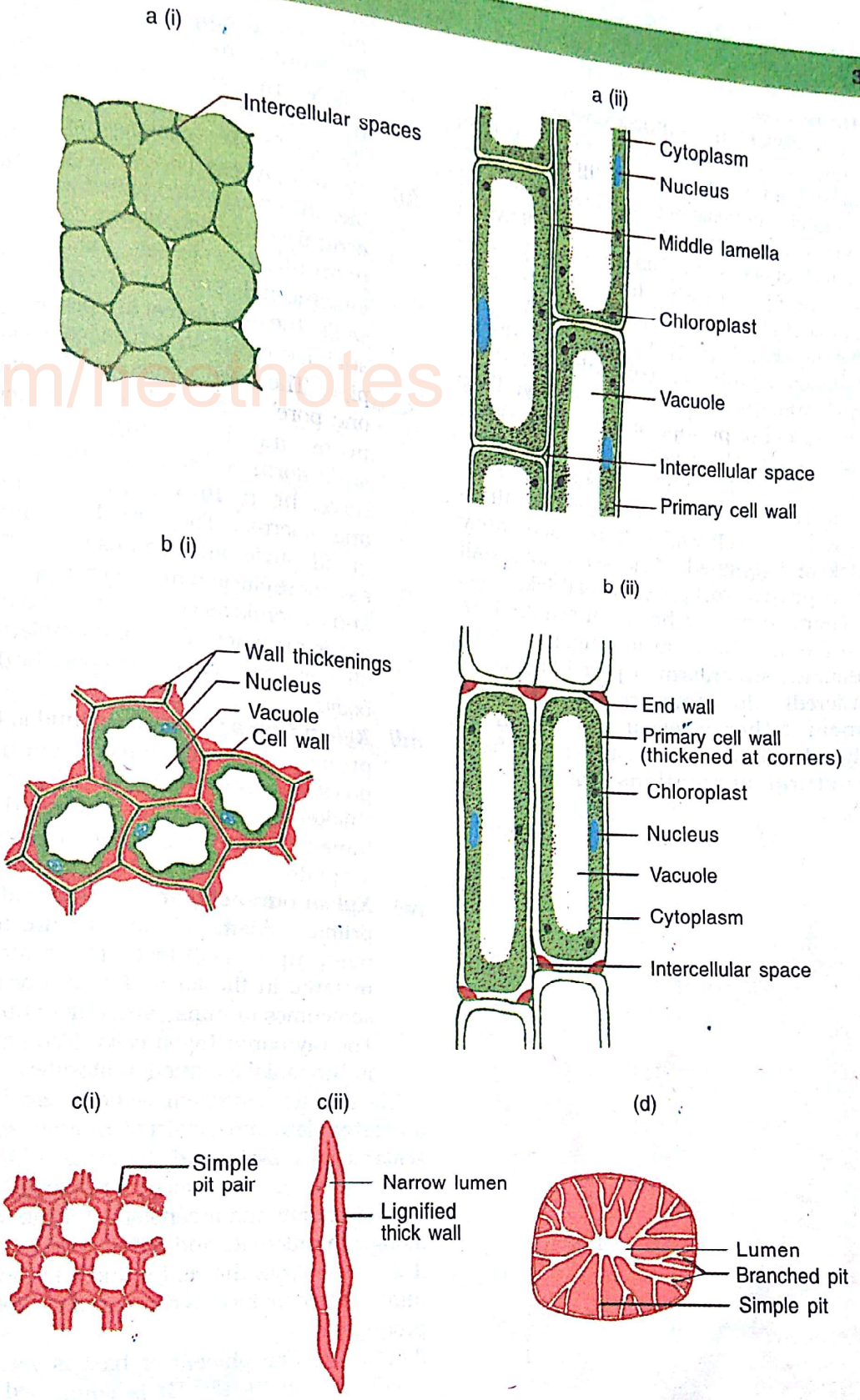

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Fig. 17.3 *Various types of simple tissues (a) Parenchyma (i) Transverse section (ii) Longitudinal section (b) Collenchyma (i) Transverse section (ii) Longitudinal section (c) Fibre (i) Transverse section (ii) Longitudinal section (d) Sclereid*

Complex tissues

Xylem and phloem are complex tissues.

(a) Xylem. It is a conducting tissue and is composed of four elements of different kinds: (i) tracheids (ii) vessels (iii) xylem fibres and (iv) xylem parenchyma. The function of xylem is to conduct water and mineral salts upwards from the root to the leaf and to give mechanical strength to the plant body.

(i) ***Tracheids*** : A single tracheid is a highly elongated or tube-like cell with hard, thick and lignified walls and a large cavity. They are devoid of protoplast and hence dead. The ends of the tracheids are tapering, blunt or chisel-like (Fig. 17.4a). These are constituents of the xylem of primitive plants. The cell wall is hard, moderately thick and lignified. The secondary wall layers possess various kinds of thickenings in them and may be distinguished as annular (in the form of rings), spiral, reticulate, scalariform or pitted (simple or bordered). In a transverse section these appear either circular, polygonal or polyhedral in outline. On account of these structural adaptations, besides the mechanical support to the plant body, these carry out transport of water, hormones and solutes from the root to the stem, leaves and the floral parts. In gymnosperms, these are the chief water transporting elements.

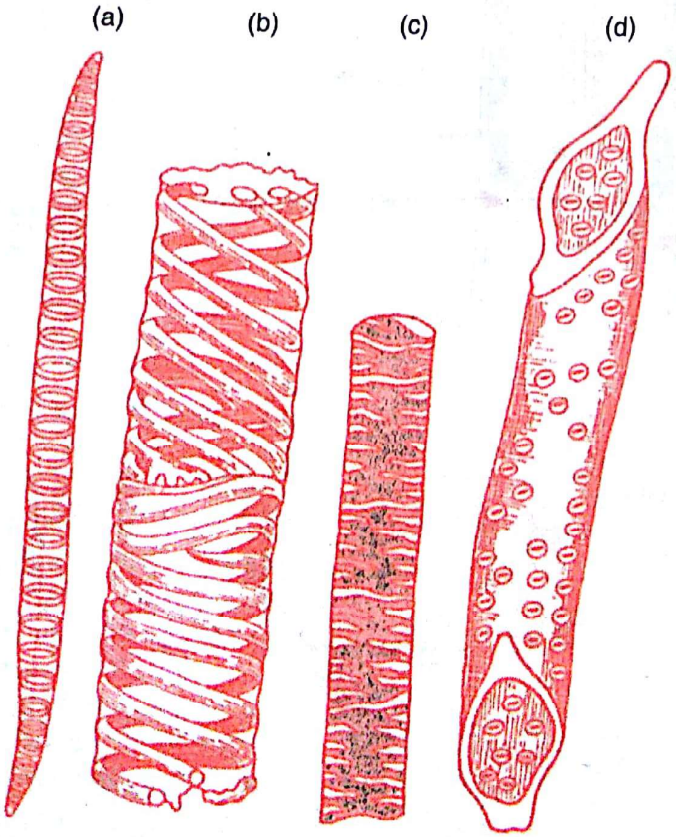

Fig. 17.4 *Different types of thickening (a) Annular (b) Spiral (c) Scalariform (d) Pitted*

(ii) ***Vessels*** : A vessel is a long, cylindrical, tube-like structure with lignified walls and a wide central cavity. The cells are dead and without protoplast. These are arranged in longitudinal series in which the transverse walls (the end plates) are perforated and as such the entire structure looks like a water pipe. The perforations may be simple (only one pore) or multiple (several pores). The pores may be arranged in reticulate, scalariform or foraminate manners. Vessels have been found in a majority of angiosperms. They are also found in a few pteridophytes and gymnosperms. They serve as a more efficient mode of transport of water and minerals as compared to tracheids due to the presence of perforation plates. These also give mechanical support to the plant body.

(iii) ***Xylem fibres*** : These are found in both the primary and secondary xylem and may possess simple or bordered pits, highly thickened walls and obliterated central lumen. These may either be septate or aseptate.

(iv) ***Xylem parenchyma*** : The cell walls of the primary xylem parenchyma are thin and made up of cellulose. These store food material in the form of starch or fat, and sometimes tannins, and other substances. The ray parenchyma cells also participate in the radial conduction of water.

The first-formed xylem elements are described as **protoxylem** and consist of annular, spiral and scalariform vessels, and lie towards the center of the stem. The latter-formed xylem is described as **metaxylem** and it consists of some tracheids alongwith reticulate and pitted vessels. In stem, it lies away from the center and its vessels have much bigger cavities as compared to those of the protoxylem.

(b) Phloem, The phloem or bast is yet another conducting tissue. It is composed of four elements: (i) sieve cells or sieve tube elements, (ii) companion cells (iii) phloem parenchyma, and (iv) phloem fibres. The main function of the phloem is to conduct prepared food

materials from the leaf to the storage organs and growing regions of the plant.

(i) ***Sieve tube elements*** : These occur as long, slender tube-like structures, arranged in longitudinal series, and associated with the companion cells (Fig. 17.5a). Their end walls are perforated (sieve pores) in a sieve like manner to form the sieve plate. These may either be simple or compound, and at maturity become impregnated with **callose**. They are devoid of nucleus at maturity. However, they possess a peripheral cytoplasm as well as a large vacuole. The uniqueness of the sieve tube is that although without nucleus, it is living and the nucleus of the companion cells control its functional activites. Distinct proteinaceous inclusions, the P-proteins (P-phloem) are seen evenly distributed throughout the lumen of the sieve tube. During wounding, alongwith callose, P-proteins help in sealing. However in lower vascular plants and gymnosperms in place of sieve tube elements, sieve cells are present. Those are narrow, elongated cells with less conspicuous sieve areas located laterally. They taper at the end or have inclined walls.

(ii) ***Companion cells*** : These are specialised parenchyma cells which are closely associated with the sieve tube elements in their origin, position and function. These originate from the same meristematic cells that give rise to the sieve tube elements (Fig. 17.5b).

The sieve tube elements and companion cells are connected by pit fields present in their longitudinal walls, which is a common wall for both and, with the death of one, the other cell also dies. The companion cells play an important role in the maintenance of a pressure gradient in the sieve tubes.

(iii) ***Phloem parenchyma*** : This is made up of elongated, tapering to broadly cylindrical, living cells which have a dense cytoplasm and nucleus. The cell wall is composed of cellulose, with pits, interconnecting axial parenchyma cells and ray cells. The phloem parenchyma stores organic food materials and other substances such as resins, mucilage, latex, etc.

(iv) ***Phloem fibres/bast fibres*** : These are much elongated, unbranched (rarely branched) and have pointed, needle-like apices. Their cell wall is quite thick with simple or slightly bordered pits. At maturity, these fibres lose

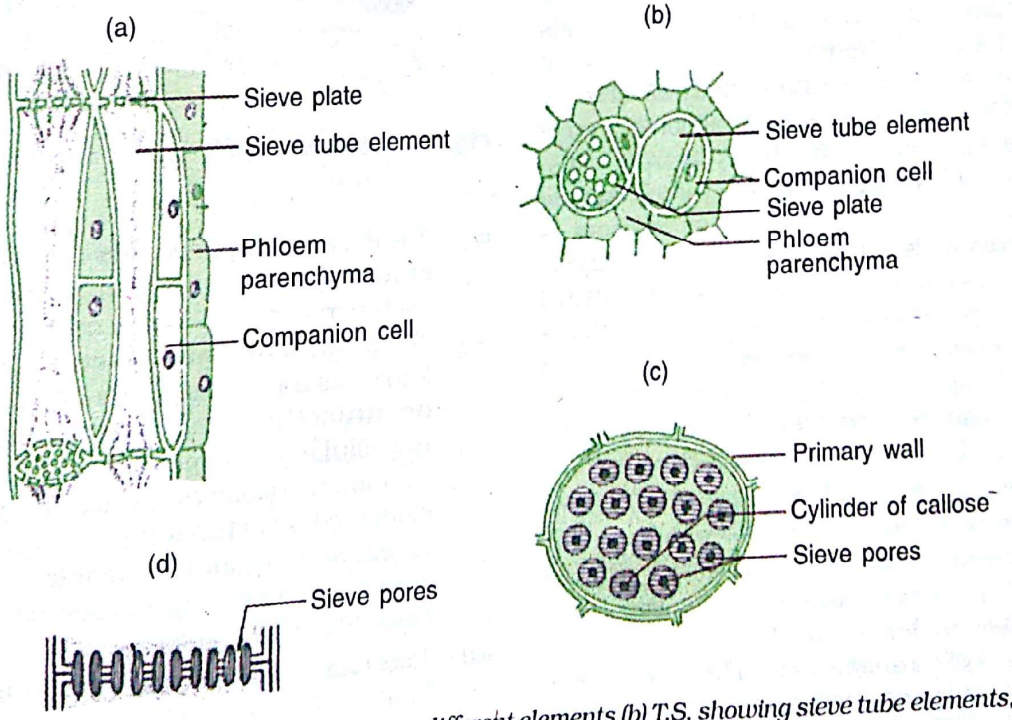

Fig. 17.5 *Structure of phloem (a) L.S. showing different elements (b) T.S. showing sieve tube elements, companion cell and phloem parenchyma (c) surface view of a sieve plate (d) L.S. of a sieve plate*

their protoplast and become dead. These occur in groups, as sheets or cylinders, e.g. in *Linum usitatissimum* (flax) and *Corchorus capsularis* (jute).

The outer portion of the phloem, consisting of narrow vessel elements constitutes the **protophloem**. The inner portion is made up of broader sieve tube elements which make the **metaphloem**.

17.2 THE TISSUE SYSTEM

It was Sachs, a German scientist, who for the first time in 1875 attempted to classify the tissues on the basis of their position and morphology. According to him, the following three categories of tissue systems can be distinctly identified: (i) the epidermal tissue system, (ii) the ground or fundamental tissue system, (iii) the vascular/stele/conducting tissue system.

(i) The epidermal tissue system comprises the following:

(a) *Epidermis* : The epidermis (*epi* : upon ; *derma* : skin) is the outermost layer of the plant body. It is made up of elongated, compactly arranged cells which constitute a continuous layer without any intercellular spaces. The cells have a large, central vacuole surrounded by a thin layer of protoplasm. The epidermis may also be multilayered as in the aerial roots of orchids and leaves of *Nerium*. The outer wall of epidermis is thick and usually covered by a **cuticle** formed by the deposition of a waxy material secreted in the epidermal cells. The cuticle is the thickest in the xerophytic plants. The outermost layer of roots is referred to as **epiblema** or **piliferous layer**. The outer walls of its cells extend outward and form tubular, unicellular prolongations called **root hair**. There are no stomata and cuticle on the epiblema. The leaves bear them abundantly usually on the lower epidermis.

(b) *Stomata* : Several minute openings or **stomata** (*sing.* stoma) are found on the epidermis of all the green aerial parts of plants, but are abundant on the lower surface of leaves as they regulate the process of transpiration. They are sunken in the pits or cavities in xerophytes. A large number of stomata occur on the upper surface of leaves of floating aquatic plants. Each stoma is surrounded by two semilunar cells known as the **guard cells**. In the dicotyledonous plants these are bean-shaped, but in sedges and grasses these are dumb-bell-shaped. The guard cells are living and possess chloroplasts. Their outer walls are thin whereas the inner ones surrounding the aperture are highly thickened. Due to this variation in the thickening, the guard cells may become turgid and flaccid, depending upon the supply of water in them, which makes the opening and closing of stomata possible. Sometimes a few neighbouring epidermal cells, in the vicinity of the guard cells become specialised in their shape, size and contents. These are known as **subsidiary accessory cells**. In such a situation, the stomatal aperture, guard cells and the surrounding subsidiary cells are together called **stomatal apparatus** (Fig. 17.6).

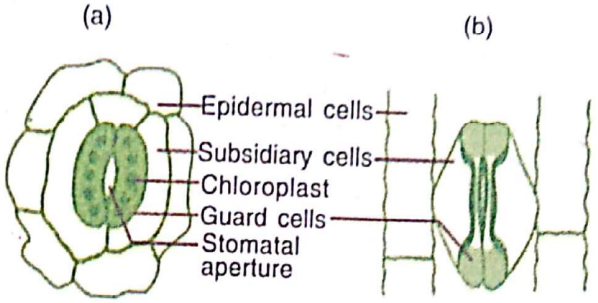

Fig. 17.6 *Stomatal apparatus (a) dicot leaf, (b) grass leaf*

c) *The epidermal appendages :* The cells of the epidermis give rise to a number of protuberances which vary markedly in their shape, structure and function. These are known as trichomes. These appendages can be unicellular or multicellular. The unicellular trichomes are usually simple, unbranched (sometimes also branched) and elongated structures (Fig. 17.7a). On the other hand, the multicellular trichomes and glands are made up of several layers of cells (Fig. 17.7b and c).

(d) *Root hair :* The epidermis of roots bears root hair in the specialised region – the root hair zone. The root hairs are formed due to the elongation of the epidermal cells and

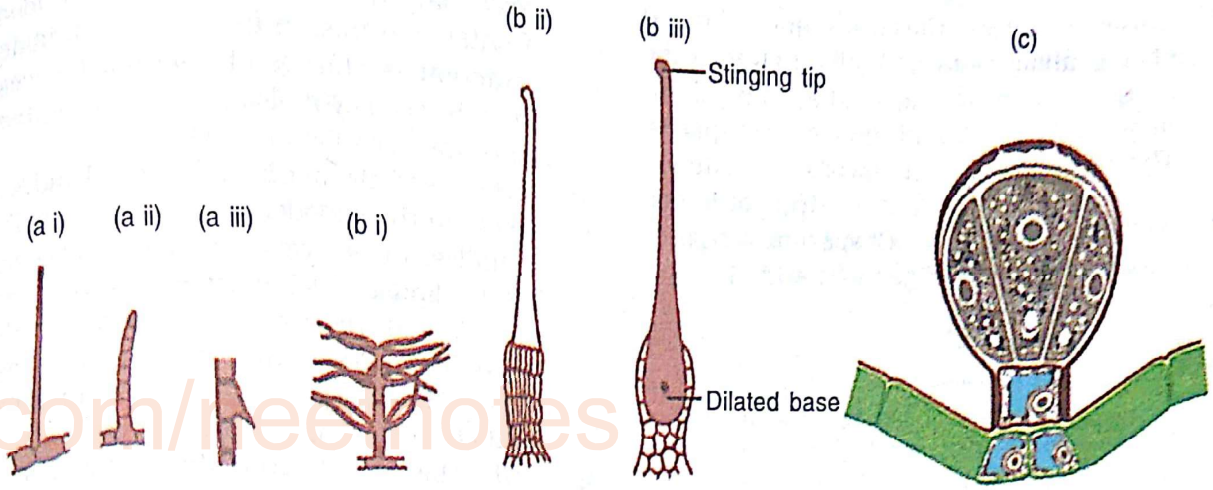

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Fig. 17.7 *Various types of trichomes (a) Simple (i) unicellular (ii) multicellular (iii) multicellular with protuberance (b) Multicellular (i) branched (ii and iii) stinging (c) glandular*

are not protuberances or appendages. These have a vacuolate protoplasm and the nucleus moves towards the apical part of the cell. The thin wall is made up of cellulose and pectic materials. The root hairs are ephemeral (short-lived) structures which play an important role in anchoring the plant body in the soil besides absorbing water and mineral solution from it.

The epidermal tissue system has the following functions: (a) On account of the presence of the cuticle, this tissue system helps in checking excessive loss of water. (b) The stomata present on the leaves help in carrying out various physiological processes such as transpiration and exchange of gases. (c) The trichomes help in protection, dispersal of seeds and fruits, and reduction of water loss.

(ii) The ground tissue system forms the main bulk of the plant body. It includes all the tissues except epidermis and vascular bundles. It is partly derived from the periblem and partly from the plerome. The primary function of this tissue system is storage and manufacture of food material. It also has a mechanical function. This system has different kinds of tissues such as parenchyma, collenchyma and sclerenchyma; of these, parenchyma is most abundant and carries out a variety of functions. Usually it is differentiated into three main zones: (a) Cortex (b) Pericycle (c) Pith and medullary rays

(a) *Cortex :* Cortex is the main zone which lies between the epidermis and pericycle and is made up of primary tissues. This zone varies in thickness from a few layers to many layers. In monocotyledonous stem the cells in this zone are predominantly parenchymatous. Similarly the cortex of root is also parenchymatous. In the leaves the ground tissue, consisting of thin walled-chlorenchyma is called **mesophyll**. On the upper epidermal side of the leaf, it is composed of palisade parenchyma, whereas on the lower epidermal side, it is differentiated into irregular or isodiametric, spongy parenchyma. In the dicotyledonous stems the cortex is usually differentiated into the following three sub-zones:
(i) hypodermis, (ii) general cortex, and (iii) endodermis.

The hypodermis is situated below the epidermis. It is few-layered thick and is made up of collenchymatous or sometimes sclerenchymatous cells. The general cortex or cortical parenchyma is few-layered thick and located below the hypodermis. It consists of thin-walled, uniform, round and spherical parenchymatous cells which have copious intercellular spaces or angular cells without such spaces. In some cases, chlorenchyma or aerenchyma may be differentiated from these cells to help in the adaptation of the plant to its specialised habitat. The endodermis is the innermost uniformly

uniseriate layer of the cortex and is formed of tangentially elongated cells. In transverse section, it appears barrel-shaped arranged in single layer without intercellular spaces. The cells show depositions of waxy material, the suberin, in the form of strips or bands which are known as **Casparian strips** or Casparian bands (Fig. 17.8a and b).

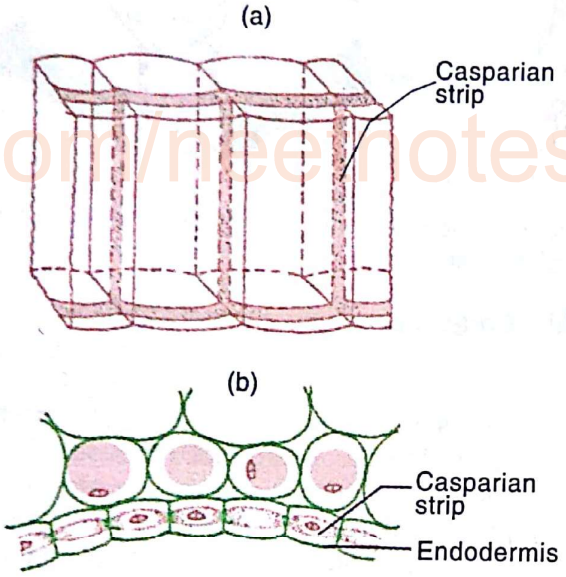

Fig. 17.8 *Endodermal cells with wall thickening or Casparian strips in two different views*

The cortex has different primary and secondary functions. In the stems, it basically serves as protective zone. Its secondary functions are storage of food. Contrary to this, in the roots the primary function is storage of food substances. Simultaneously it also helps in absorption and translocation of water.

(b) *Pericycle* : This zone is multilayered and lies between the endodermis and the vascular bundles. In dicotyledonous stems it occurs as a cylinder which encircles the vascular bundle and the pith. It is generally made up of both parenchyma and sclerenchyma (Fig. 17.9). The pericycle is absent in roots and stems of some aquatic plants.

(c) *Pith* : The central part of the ground tissue is known as pith or medulla. Usually this is made up of thin-walled, parenchymatous cells which may be with or without intercellular spaces. The dicotyledonous stems have a large and distinct pith which also extends in between the vascular bundles in the form of narrow parenchymatous structure known as **medullary rays**. The pith forms the central core of stems and roots. It chiefly serves as the store house of several excretory substances, such as tannins, phenols, calcium oxalate, etc.

(iii) The vascular tissue system is formed by the phloem and xylem. The elements of xylem (tracheids, vessels, xylem fibres and xylem parenchyma) and phloem (sieve cells or sieve tube elements, companion

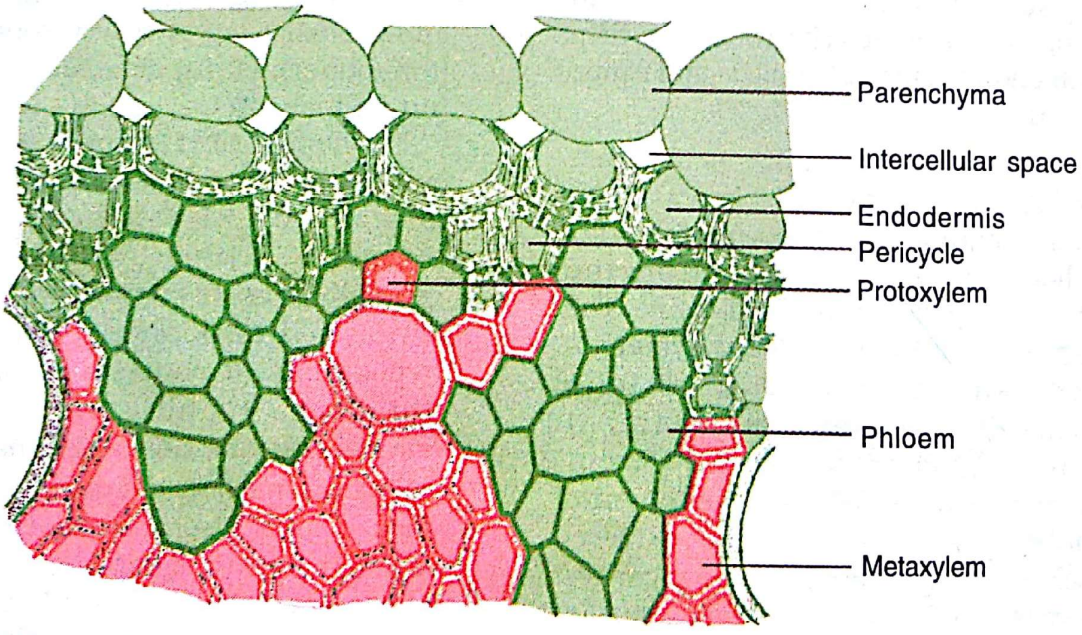

Fig. 17.9 *T.S. of a portion of maize root showing position of endodermis and pericycle*

cells, phloem parenchyma and phloem fibres/ bast fibres) are always organised in groups and are called **vascular bundles**. In the open vascular bundles of dicotyledons, a meristematic region, **cambium** is present between the phloem and xylem which is known as the **intrafascicular cambium**. Contrary to this, in the monocotyledons, the vascular bundles are without cambium and are called closed.

Depending upon the organs of plants and their function in the plant body, the arrangement of xylem and phloem differs. Accordingly, the vascular bundles are of the following types:

(i) **Radial** vascular bundles have xylem and phloem which are arranged in an alternate manner on different radii. Such bundles are mainly found in the roots;

(ii) **Conjoint** vascular bundles have xylem and phloem situated at the same radius and form a vascular bundle together (Fig. 17.10). These are usually found in stems and leaves.

Depending upon the mutual relationship of xylem and phloem, these are divided into three types.

(a) Collateral: When the phloem is located only on the outside to the xylem. This can either be open or close as described earlier;

(b) Bicollateral: In the vascular bundles of this category, the phloem is found in two groups, one outside the xylem elements and the other inner to them. These are characteristically found in the stems of members of the family Cucurbitaceae. Due to the presence of the cambium, these are always open as in pumpkin (*Cucurbita pepo*) and ridge-gourd (*Luffa cylindrica*); and

(c) Concentric: The bundle, in which either phloem surrounds the xylem or xylem surrounds the phloem completely, is known as concentric. This is of two types:

(i) Amphicribral: xylem lies at the centre and is surrounded by a ring of phloem, e.g. fern;

(ii) Amphivasal : phloem lies at the centre and is surrounded by the xylem, e.g. *Dracaena*. (Fig. 17.10 e,f).

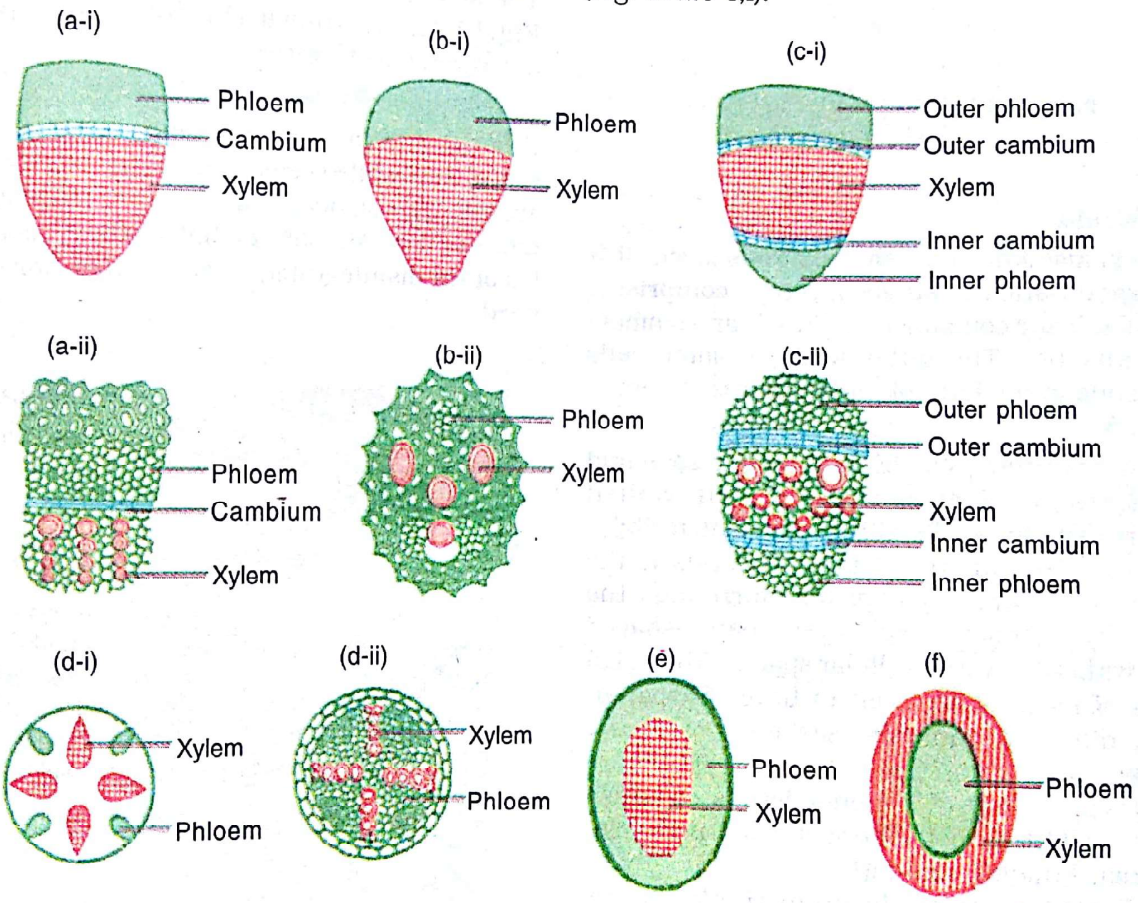

Fig. 17.10 *Various types of vascular bundles (a) Conjoint, collateral and open vascular bundle (i and ii) (b) conjoint collateral and closed vascular bundle (i and ii), (c) Conjoint, bicollateral and open vascular bundle (i and ii), (d) Radial vascular bundle (i and ii), (e) Amphicribral, (f) Amphivasal*

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17.3 INTERNAL STRUCTURE OF DICOT AND MONOCOT PLANTS

Dicotyledonous Root

The primary structure of a typical dicotyledonous root can be studied by examining the transverse section of a young root of sunflower, pea or gram (Fig. 17.11). It shows the following plan of arrangement of tissues:

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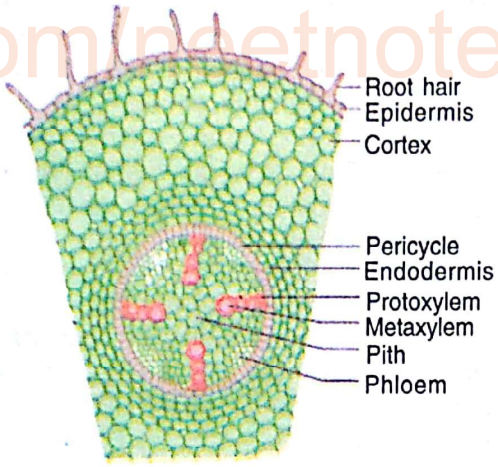

Fig. 17.11 T.S. of root of sunflower (Helianthus annuus)

Epiblema

This is also known as the piliferous layer. It is characteristically single layered, comprising tubular living components. Cuticle and stomata are absent. The outer walls of some cells protrude in the form of unicellular root hair.

Cortex

It is a relatively simple and massive zone and consists of several layers of thin-walled parenchymatous cells with plenty of intercellular spaces. The innermost layer of cortex is the endodermis which completely surrounds the stele. It comprises a single layer of barrel-shaped cells without any intercellular spaces. The radial walls of its cells are seen to have Casparian thickening in a transverse section.

Stele

All tissues inside of the endodermis comprise the stele of the root. These include pericycle, vascular bundles, and pith:

(i) Pericycle is made up of thick-walled parenchymatous cells. The pericycle is the seat of origin of lateral roots or the root branches;

(ii) Vascular bundles are radial, that is, xylem and phloem which occur in separate patches, are arranged on alternate radii. Xylem consists of both proto and metaxylem. It is exarch since the protoxylem lies towards the epidermis. The number of xylem bundles and also the phloem in dicot roots may be two to six, i.e. diarch to hexarch (exceptionally it may be triarch as in roots of pea). In some species, the metaxylem of all xylem bundles may meet in the centre to form a xylem plate due to which the pith is obliterated. Phloem patches are rather small. They form oval groups under the pericycle. These oval masses are separated from the xylem by parenchymatous cells that are known as the **conjunctive tissue**.

(iii) Pith is generally absent in dicotyledonous roots and, if present, it is small.

Monocot Root

The structure of a monocot root is shown in Fig.17.12. It shows the following plan of arrangement of tissues:

Epiblema or Piliferous Layer

It is the outermost layer consisting of a single row of thin-walled cells without any intercellular spaces. The structure and fate of this layer is more or less similar to that of dicot roots. It becomes disintegrated after the root hairs are shed.

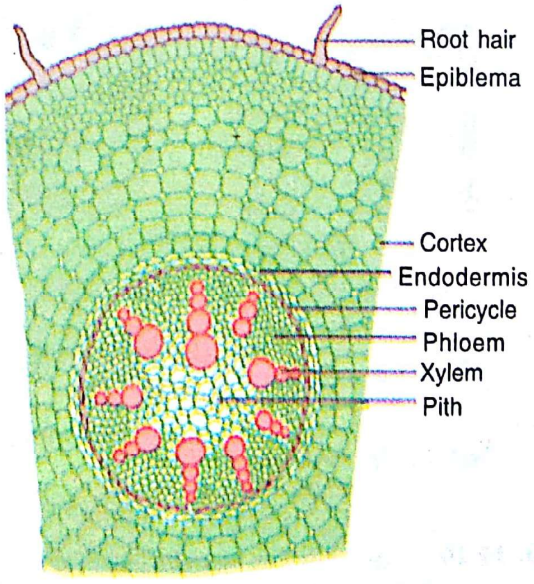

Fig. 17.12 T.S. of a typical monocot root

Cortex

The cortex consists of a large parenchymatous zone having oval or spherical cells with intercellular spaces. The cells are colourless and store water. As the epiblema dies off, a few outer layers of the cortex become cutinised and form the exodermis. The innermost layer of the cortex is **endodermis**. This forms a complete ring around the stele. Its cells are barrel-shaped. Their radial walls are thickened due to the formation of Casparian strips and deposition of suberin over them. Certain endodermal cells which are present opposite to the xylem bundles remain thin-walled. These are known as **passage cells**. Their function is to transfer water and dissolved salts from cortex directly into the xylem.

Stele

All tissues inside of the endodermis comprise the stele. These include pericycle, vascular bundles and pith:

(i) Pericycle - This is the outermost layer of the stele and lies just below the endodermis. It consists of one or a few layers of parenchymatous cells. It gives rise to the lateral roots;

(ii) Vascular bundles - Monocot roots are usually polyarch. The xylem and phloem form an equal number of separate bundles and they are arranged in a ring. The arrangement is radial. The xylem is exarch. The parenchyma is present in between the xylem and phloem bundles. These cells are called conjunctive tissue;

(iii) Pith is very well developed and parenchymatous.

Table 17.1 presents differences between dicotyledonous and monocotyledonous roots.

Table 17.1 Differences between Dicotyledonous and Monocotyledonous Roots

Features	Dicotyledonous Root	Monocotyledonous Root
Xylem bundles	vary from 2 to 6	usually more than six (polyarch)
Pith	small or lacking	very large and well developed
Secondary growth	takes place	does not take place

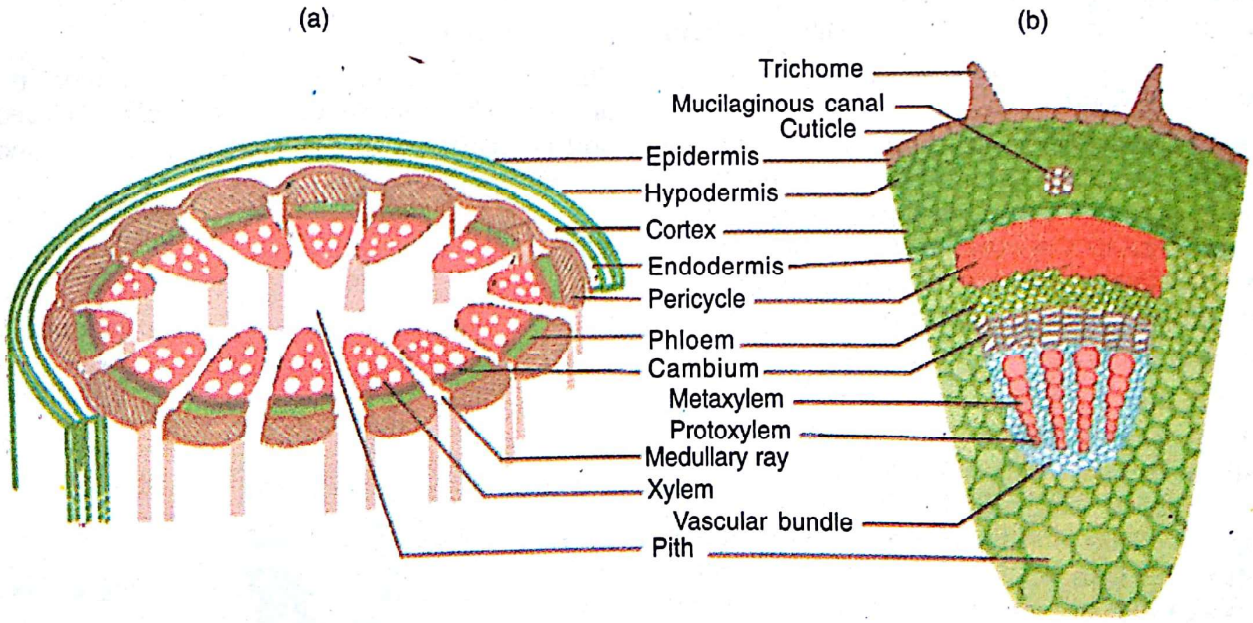

Fig. 17.13 T.S. of stem of sunflower (*Helianthus annuus*) (a) Outline diagram (b) Cellular enlarged portion

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Dicotyledonous Stem

The transverse section of the young sunflower (*Helianthus annuus*) stem shows the following structure (Fig. 17.13).

Epidermis

This is the outermost layer of the stem. It consists of a single layer of cells. The epidermis bears multicellular, uniseriate trichomes. A thin layer of cuticle is present on the epidermis as well as the trichomes.

Cortex

The cortex consists of several layers of cells. This is divided into three sub-zones – hypodermis, general cortex and endodermis: (i) Hypodermis – This is just below the epidermis and consists of 3 to 4 layers of collenchymatous cells. It gives mechanical strength to the stem. These cells are thickened at the corners and possess chloroplasts; (ii) General cortex – The cells of this zone are parenchymatous and multilayered. Secretory (Oil) ducts, surrounded by a glandular parenchy- matous layer, also occur copiously throughout this sub-zone; (iii) Endodermis – It is the innermost layer of the cortex, and consists of a single layer of barrel-shaped cells. Since it is rich in starch, it is also referred to as the **starch sheath.**

Pericycle

It is in the form of semilunar patches of sclerenchyma. Each patch associated with phloem of the vascular bundle, is called the **hard bast.**

Medullary Rays

In between the vascular bundles there are a few layers of parenchymatous cells which constitute medullary rays.. These are slightly larger in size as compared to the other cortical cells. Usually these are polygonal in outline and exhibit no intercellular spaces in between.

Vascular Bundles

The vascular bundles are arranged in a ring internal to the endodermis. Each vascular bundle is conjoint, collateral, endarch and open. It is composed of xylem, phloem and cambium: *(i) Phloem :* This is situated on the outerside of the vascular bundle. The cells are thin-walled and polygonal. Phloem is a complex tissue and is composed of sieve tube elements, companion cells, the phloem parenchyma and phloem fibres; *(ii) Xylem :* The xylem tissue lies below the phloem. This is composed of the vessels, tracheids, xylem parenchyma, and the fibres; *(iii) Cambium :* This is present in between the xylem and the phloem. It consists of 2-3 layers of thin-walled, rectangualr cells.

Pith

This is the central portion of the stem. It consists of rounded, parenchymatous cells with plenty of intercellular spaces.

Monocotyledonous Stem

The internal structure of the young maize (*Zea mays*) stem, which is a monocot shows the following details in a transverse section (Fig. 17.14).

Epidermis

This is the outermost layer of cells which is usually single layered. It is covered with a thick cuticle and bears a few stomata.

Hypodermis

The cells of hypodermis are sclerenchymatous. This layer is located just below the epidermis.

Ground tissue

The entire tissue lying inside the epidermis, except for the vascular bundles, is known as the ground tissue. It consists of rounded parenchymatous cells with distinct intercellular spaces. The ground tissue is not differentiated into cortex, endodermis, pericycle and pith.

Vascular bundles

The vascular bundles are many in number and are of variable size. They are collateral and closed and lie scattered in the ground tissue. Each

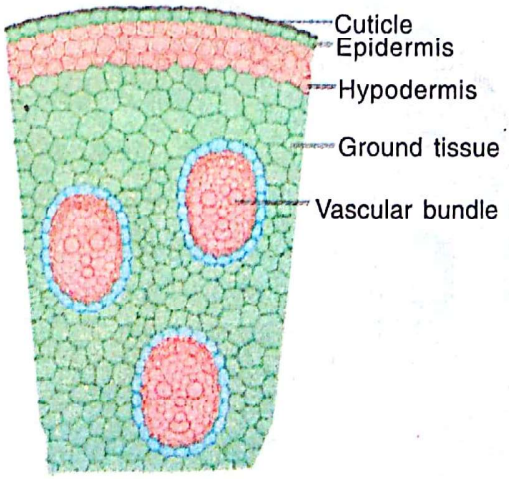

Fig. 17.14 *T.S. of stem of maize (Zea mays)*

Table 17.2 Major Anatomical Differences between Dicotyledonous and Monocotyledonous Stems

Features	Dicotyledonous stem	Monocotyledonous stem
Trichomes	present on the epidermal layer	usually absent
Hypodermis	collenchymatous	sclerenchymatous
Cortex	a few layers of parenchyma	a continuous mass of parenchyma
Vascular bundles	arranged in a ring	scattered in the ground tissue
	not surrounded by any bundle sheath	surrounded by a sclerenchymatous bundle sheath
	collateral and open	collateral and closed
	phloem parenchyma present	phloem parenchyma absent
	usually wedge shaped	usually oval shaped
	the water containing cavities are absent.	the water containing cavities are present

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vascular bundle is oval and usually surrounded by a sheath of sclerenchymatous cells, the **bundle sheath.** The bundle consists of two parts, xylem and phloem:

(i) The xylem consists of 3-4 distinct vessels which are arranged in the form of a 'Y'. One or two smaller vessels lying at the arm base constitute the protoxylem. The two bigger vessels lying laterally constitute the metaxylem. The lowermost protoxylem vessels open into a cavity called water containing cavity;

(ii) The phloem lies outer to the xylem. It consists of sieve tube elements and companion cells. Phloem parenchyma is absent.

Table 17.2 summarises major differences in the internal structure of dicotyledonous and monocotyledonous stems.

Dicotyledonous (Dorsiventral) Leaf

The vertical section of a dorsiventral leaf shows the following features (Fig. 17.15).

Epidermis

Upper and lower surfaces of leaf are bound by the epidermis, known as the upper and lower epidermis, respectively. The upper epidermis generally comprises a single layer of cells.

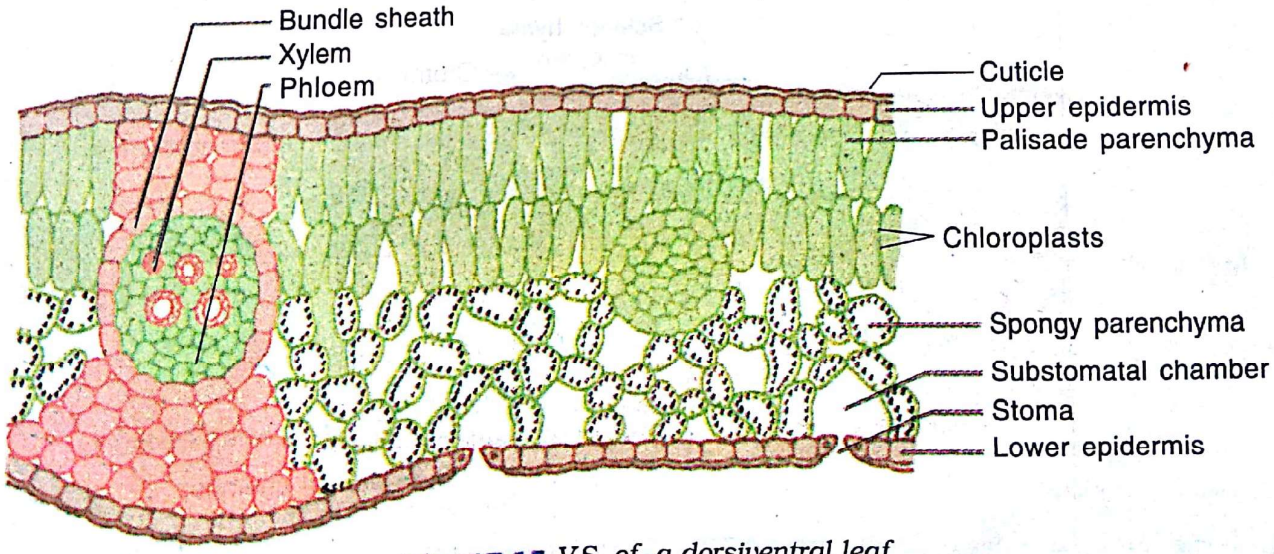

Fig. 17.15 V.S. of a dorsiventral leaf

A thick cuticle is usually present on the outer walls of these cells. The stomata are absent and if present, these are fewer in number as compared to those on the lower epidermis. The lower epidermis is also made up of a single layer of cells bearing a large number of stomata and is covered by a thin layer of cuticle. Each stoma is surrounded by two guard cells which contain chloroplasts and open into a large space below, called the **Sub-stomatal chamber**. On account of the presence of chloroplasts, they are able to carry out photosynthesis to a limited extent.

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Mesophyll

The entire tissue between the upper and lower epidermis is called the mesophyll. The mesophyll consists of two regions; the palisade parenchyma and the spongy parenchyma. The palisade parenchyma are vertically elongated cells which may be organised in one or more layers. The palisade cells are compactly arranged through out and show only little intercellular spaces. They possess abundant chloroplasts and a large vacuole. Due to the presence of chloroplasts, the palisade cells carry out photosynthesis in the presence of sunlight. The spongy parenchyma are components of the remaining portion of the mesophyll tissue which is extended from below the palisade layer to the lower epidermis. Its cells are oval or rounded, enclosing numerous large air spaces and air cavities. These spaces open towards the outside through the stomata and help in the diffusion of gases. Spongy parenchyma cells contain chloroplasts to perform photosynthesis, like those of the palisade parenchyma.

Vascular Bundles

The vascular bundles are conjoint, collateral and endarch. Each vascular bundle is surrounded by a layer of thick-walled cells arranged compactly and known as the **bundle sheath**. The xylem is situated towards the upper epidermis (adaxial surface) and phloem lies towards the lower epidermis (abaxial surface).

Monocotyledonous (Isobilateral) Leaf

A section of monocotyledonous leaf reveals the following internal structure (Fig. 17.16).

Epidermis

This is the outermost covering of the lamina. It is a single layer of thin-walled, barrel-shaped, non-green cells on both the lower and upper surfaces. It is cuticularized and possesses more or less equally distributed stomata on both the surfaces. The guard cells of the stomata contain chloroplasts. In grasses, a few large, empty and colourless cells are also present at intervals on the upper surface. These are called the **bulliform cells**.

Mesophyll

It is not differentiated into palisade and spongy parenchyma. All the cells are thin-walled and parenchymatous. The cells are circular and uniform and contain numerous chloroplasts. These are compactly arranged and have only a few intercellular spaces.

Vascular Bundles

Many vascular bundles can be observed in the leaf, arranged in a parallel manner. Each vascular bundle is conjoint, collateral, endarch and closed, and is surrounded by a

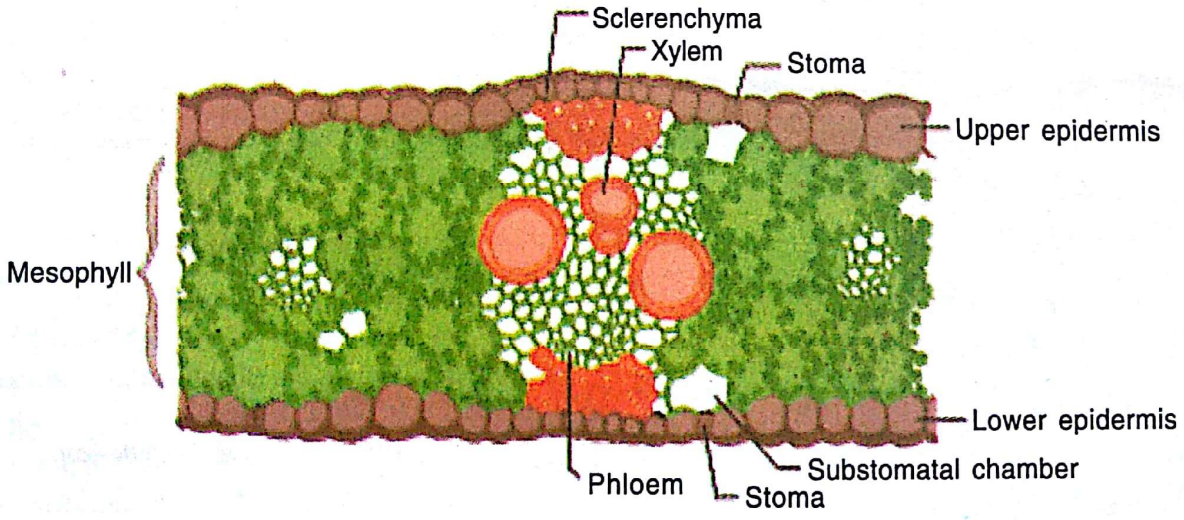

Fig. 17.16 *V.S. of an isobilateral leaf*

Table 17.3 Major Anatomical Differences between Dorsiventral and Isobilateral Leaf

Features	Dorsiventral leaf	Isobilateral leaf
Cuticle	Thick at upper epidermis and thin at lower epidermis	Uniform cuticle
Stomata	More on lower surface	Equal number of stomata on either side
Mesophyll	Differentiated into palisade and spongy parenchyma	Not differentiated into palisade and spongy parenchyma

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distinct parenchymatous bundle sheath in some grasses. Two distinct patches of sclerenchyma are present above and below each of the large vascular bundles and extend up to the upper and lower epidermal layers, respectively. The xylem is located towards the upper epidermis and the phloem towards the lower epidermis (endarch).

Table 17.3 includes major anatomical differences between dorsiventral and isobilateral leaf.

17.4 SECONDARY GROWTH

In most dicotyledonous stems and roots, distinct secondary growth is visible, which increases the diameter of the stems and roots (Fig. 17.17). The salient features of this process are given below:

Dicotyledonous Stem

The processes and structures associated with the secondary growth in dicot stems are given below.

Formation of the Cambial Ring

You have studied that in dicot stems an **intrafascicular cambium** is present between xylem and phloem, which is primary in nature. The parenchymatous cells of each medullary ray, lying between the intrafascicular cambium of the adjacent bundles divide and become meristematic and form a strip of cambium called the interfascicular cambium. Both types of cambia (intrafascicular and interfascicular) join

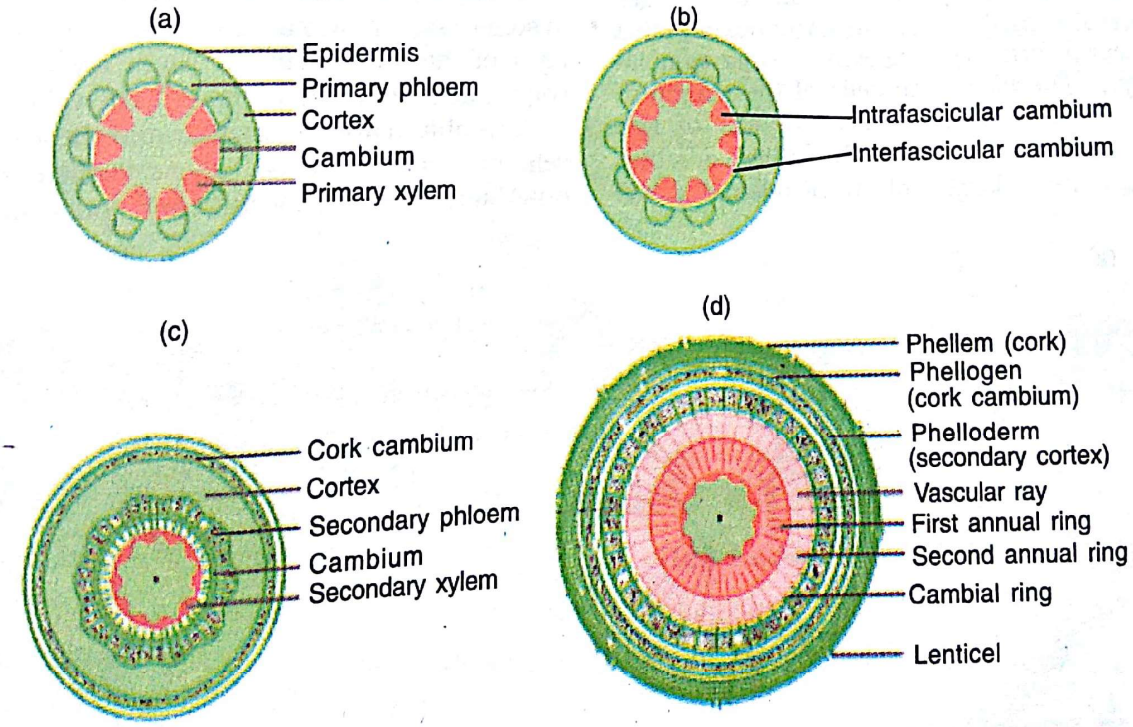

Fig. 17.17 *Schematic representation of different stages in the secondary growth in a typical dicot stem*

together to form a complete ring of vascular cambium, which is, thus partly primary and partly secondary in origin.

Activity of the Cambial Ring

The cambial ring becomes active and begins to cut off new cells, both towards the inner and outer side, by vertical or oblique divisions of the elongated fusiform initials. The derivatives which are cut off towards the outer side get differentiated into the secondary phloem whereas those produced on the inner side are differentiated into secondary xylem (Fig. 17.18). The cambium is generally more active on the inner side than on the outer side. As a result, the xylem increases more rapidly in bulk than the phloem, and soon forms a compact mass. This forms the main bulk of the plant body. Due to the continued formation of secondary xylem both the primary and secondary phloem of the earlier years get gradually crushed.

At places, cambium forms some narrow bands of parenchyma which pass through secondary phloem and the secondary xylem. These are the secondary rays.

Annual Rings

The activity of the cambium is under the control of a series of physiological and environmental factors. For example, during the spring and summer, the temperature is high which alongwith the higher relative humidity, longer duration of sunshine, and the hormones supplied by the newly formed young leaves, favour cambial activity. Therefore, the cells of the cambium divide rapidly and several layers of the undifferentiated cells become visible. Consequently, a larger volume of xylem tissue is produced, having comparatively larger, thin-walled and light-staining components. On the other hand, during winter/autumn, the temperature is low due to which the activity of the cambium also slows down. Therefore, the amount of xylem elements produced and their diameter is much less, since these are small, have thicker walls and take darker stain. The wood formed during the spring and summer is known as the **early** or **spring wood** and that produced during winter is known as **late** or **autumn wood**. The spring wood is lighter in colour and exhibits low density whereas the autumn (or winter) wood is darker and has higher density. One light and one dark coloured zone comprises one year's growth and this is known as the **annual** ring or **growth** ring. Since each annual ring corresponds to the growth of wood of one year, one can estimate the age of a tree to some degree of accuracy by counting them.

Activity of Cork Cambium or Phellogen

Due to the addition of secondary phloem and secondary xylem elements, the outermost layer of the cortex becomes highly stretched and may crack open. During this process, a few layers of meristematic tissue arise in the cortex. This is called the **cork cambium** or **phellogen**. The nature of this cambium is secondary. Although it usually arises in the outer layers of the cortex, in some cases, it may also arise in the epidermal layer of the stem or in the inner layers of the cortex, or even in the pericycle. The cells of the cork cambium are rectangular and these cut off cells on both the sides. Those formed on the outer side become suberised and constitute the

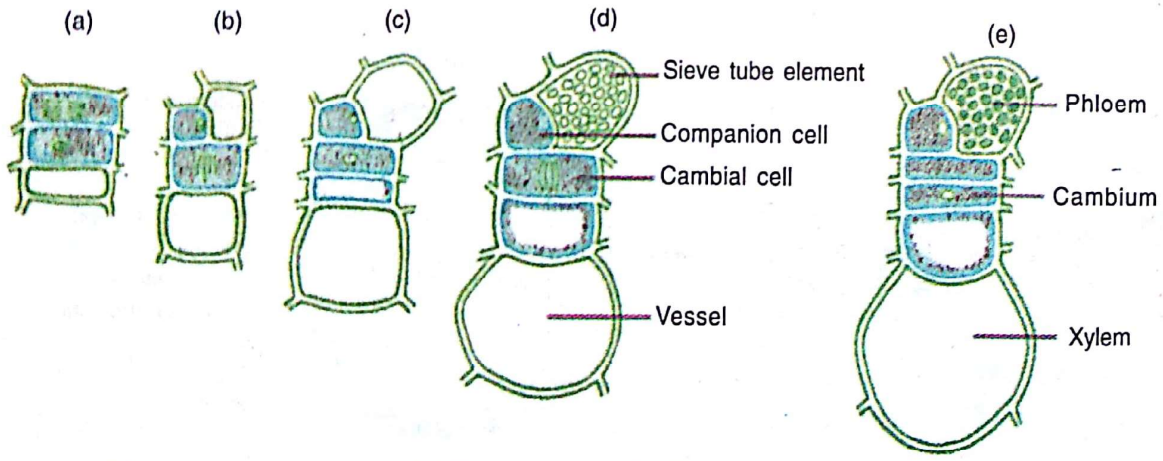

Fig. 17.18 *Schematic representation of different stages of differentiation of conducting tissues (xylem and phloem) from a cambial cell*

Cork or **phellem** which is impervious to water and air. The inner cells become parenchymatous and may contain chloroplasts. These constitute the **secondary cortex** or **phelloderm.** The cork (phellem), cork cambium (phellogen) and the secondary cortex (phelloderm) are collectively known as the **periderm.** Generally, yet another tissue, the **bark**, also becomes distinguishable in the vicinity of the periderm which includes all the tissues outside the vascular cambium, i.e. secondary phloem, the elements of the primary phloem and primary as well as the secondary cortex, in the crushed state. Its cells are living and some of them take part in the conduction of the metabolites. The commercial cork is obtained from the plant *Quercus suber*, which is commonly found in Portugal and parts of Spain.

Lenticels

These are openings or breaks in the cork tissue which look like raised spots on the surface of the stem and permit the exchange of gases between the outer atmosphere and the internal tissue of the cells of the stem. These occur in most of the woody trees except the climbers. A transverse section of the stem passing through a lenticel shows its internal structure (Fig. 17.19). Usually it consists of a pore formed due to the rupture in the epidermal layer. Below the pore is visible a loose mass of thin-walled, rounded parenchymatous cells, which is known as the **complementary tissue.** In some cases, e.g. *Prunus*, some of its cells become suberized. The number of lenticels formed in a stem is variable. They may remain scattered or become arranged in longitudinal or vertical rows. Rows of lenticels may occur opposite the medullary rays, thus facilitating free exchange of gases.

Heartwood and Sapwood

After certain years of growth, the xylem elements of the stems of a number of trees develop dark brown coloration, especially in the central or innermost layers. This region comprises dead elements with highly lignified walls and is called **heartwood** or **duramen** as against the peripheral, light coloured, living wood, which is referred to as **sapwood** or **alburnum**. The heartwood contains organic compounds like oils, aromatic substances, gums, resins, tannins, phenols, and other coloured materials, especially the essential oils. On account of these cumulative features, it is more durable and resistant to the attack of the micro-organisms and insects as compared to the sapwood.

Dicotyledonous Root

The secondary growth in a dicot root is similar to that in the dicot stem, but there is a marked difference in the manner of the formation of cambium and tissue differentiation.

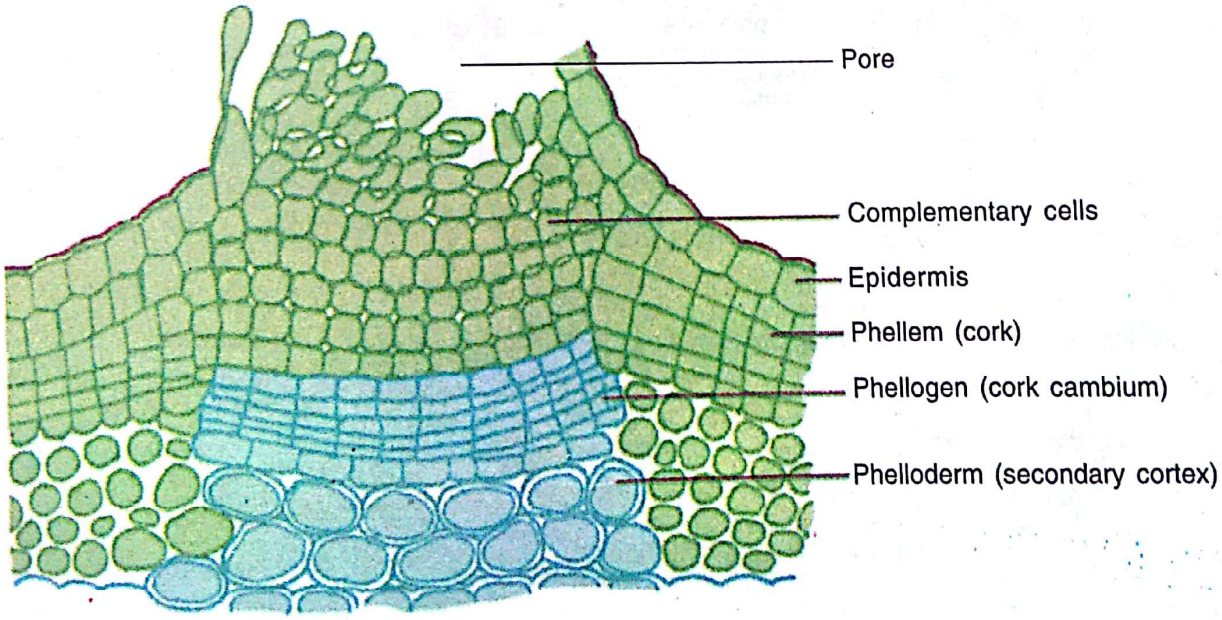

Fig. 17.19 *Structure of a lenticel*

Formation of the Vascular Cambium

The vascular cambium in a dicot root is completely secondary in origin. In the beginning, a portion of the conjunctive tissue just below the phloem bundles becomes meristematic and gives rise to brick - shaped cells of the cambium. This strip of cambium extends laterally between the xylem and phloem bundles (Fig. 17.20). At the same time, a portion of the pericycle above the protoxylem also becomes meristematic and forms a strip of cambium. Both the cambial strips join together forming a complete and continuous wavy ring which passes below the phloem bundles and above the xylem bundles.

Activity of the Vascular Cambium

The cambial ring below the phloem bundles cuts off more derivatives towards the inner side. Due to this over production of secondary tissue at this site, the cambium as well as the primary phloem are pushed outwards. As a result, the wavy band of cambium becomes circular. Now, this entire circular ring of cambium becomes active and cuts off secondary tissues on either side. The cells which are formed on the inner side of the cambium ring are differentiated into elements of secondary xylem and consist mainly of vessels, xylem parenchyma and a few fibres. The secondary tissues corresponding to each protoxylem bundle modify into parenchyma to form the primary medullary rays. These are more prominent in roots than in stems. The cambium ring on its outer side cuts off new cells which get differentiated into secondary phloem having sieve tube elements, companion cells, phloem parenchyma and phloem fibres. These together constitute the secondary phloem. The primary phloem bundles are gradually crushed and are not seen in older roots.

Formation and Activity of the Cork Cambium

The cells of the pericycle become meristematic and give rise to a few layers of thin- walled rectangular cells. These constitute the **Cork cambium** or **phellogen**. It gives rise to secondary tissue on the outer side and forms the **cork** or **phellem**. The cells, which are formed towards the inner side of the cork cambium, constitute the secondary cortex or phelloderm. During this process the endodermis, general cortex and epiblema get disorganised and gradually disappear. Lenticels may also be present in the roots scattered here and there.

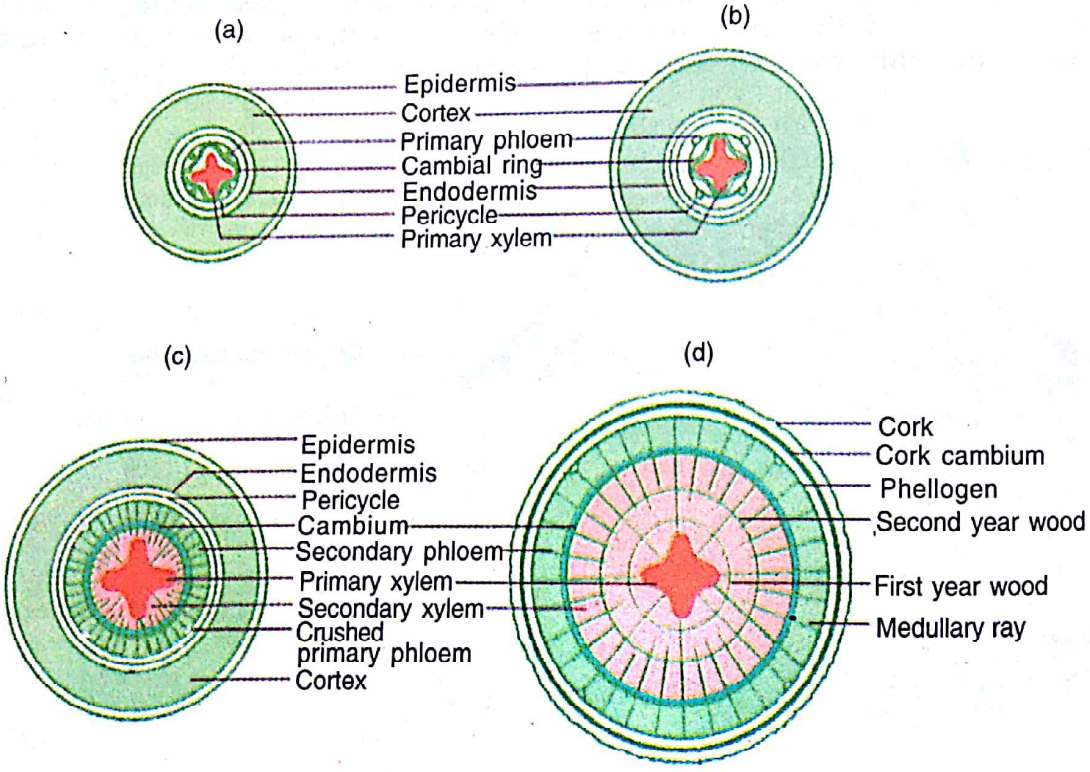

Fig. 17.20 *Diagrammatic representation of different stages of secondary growth in a typical dicot root*

SUMMARY

The plant tissues are of two types: meristematic, in which the cells are capable of division and addition of new cells, and permanent, where the cells have lost this property. On the basis of their position in the plant body, the meristematic tissues in an angiospermous plant are classified into apical, lateral and intercalary meristem. In the apical meristem lies a group of cells – the pro-meristem which is differentiated into three regions: dermatogen, periblem and plerome. The permanent tissues are of two types: (a) Simple and (b) Complex. The first category includes parenchyma, collenchyma and sclerenchyma. The parenchyma comprises variously shaped, thin-walled cells with conspicuous vacuoles, and chiefly carries out the function of storage of food substances. The collenchyma is characterised by cells which have thicker walls at their corners. It provides mechanical support to the growing organs. The sclerenchyma is distinguished by uniformly thick-walled, dead cells with a narrow lumen. This is the chief mechanical support-providing tissue, and its components are of two types, the fibres – narrow and tapering, usually occurring in groups and sclereids, short and cylindrical or polyhedral. The xylem and phloem are the complex tissues of the vascular plants. The former is made up of four types of elements: the tracheids, vessels, fibres and xylem parenchyma, whereas the latter comprises sieve cells or, sieve tube elements, companion cells, phloem parenchyma and phloem fibres.

Sachs divided the tissue system in plants into three categories: (i) Epidermal (ii) Ground tissue, and (iii) the Vascular tissue system. The epidermal tissue system is made up of epidermal cells which are thin-walled with no intercellular spaces. Its main function is to protect the internal tissues. Some minute apertures are found in the epidermis. These are called stomata and help in transpiration. There are some protuberances on the epidermal cells of the leaves and stems, called trichomes. The epidermal cells of roots elongate to form the root hair. These also increase the surface area of roots to absorb water and minerals, and furnish excellent mode of anchorage.

The ground tissue system is differentiated into three zones: (a) Cortex, (b) Pericycle, and (c) Pith and medullary rays. The cortex is differentiated into hypodermis, comprising a few, thick-walled cells, general cortex, thin-walled parenchymatous cells with copious intercellular spaces. A well-defined ring of compact cells, the endodermis forms the innermost layer of the cortex made up of an uniseriate row of vertically elongated elements with Casparian strips. The pericycle lies between the endodermis and the vascular bundles, usually made up of parenchyma and sclerenchymatous cells. It provides mechanical support and is the seat of origin of the lateral roots. The pith is parenchymatous and lies in the central/ ground tissue and stores the food materials. The vascular tissue system is formed by the phloem and xylem. Depending upon the presence or absence of the cambium, the vascular bundles are described as open or closed. If the xylem and phloem are present on different radii, the vascular bundle is stated to be radial; if these are present on the same radius, these are described as conjoint.

The internal structure of dicotyledonous root shows epidermis, a single layer with root hair, no intercellular spaces, and the cortex, a massive parenchymatous tissue made of thin-walled cells with intercellular spaces. The vascular bundles are radial, 2-6 in number, exarch, and pith either absent or very small. On the contrary in the monocotyledons, the vascular bundles are more than 6 and the pith is large.

In a dicotyledonous stem, the epidermis is single layered with multicellular trichomes, covered with cuticle. Here, the cortex is divided into collenchymatous hypodermis, a parenchymatous general cortex which may be glandular, and an uniseriate endodermis. The vascular bundles are conjoint, collateral, endarch and open. The phloem is situated on the outer side and xylem on the inner side of the vascular bundle. The cambium lies between xylem and phloem. The vascular bundles are separated from each other by medullary rays. The pith is parenchymatous with distinct intercellular spaces. In a monocotyledonous stem, the epidermis is single-layered, covered with a thick cuticle. The ground tissue is not

differentiated into cortex ,endodermis, pericycle or the pith. The vascular bundles are scattered in the ground tissue, covered with a bundle sheath. The phloem lies outer to the xylem. The pith is made up of parenchymatous cells which may get disorganised to form a lumen, in the centre.

In a dicotyledonous leaf, the upper and lower surfaces are covered by epidermis. The lower epidermis is interrupted by a large number of stomata. Between the two epidermis layers, is situated the mesophyll tissue comprising palisade and spongy parenchyma, the cells which possess abundant chloroplasts and are chiefly responsible for photosynthesis.

In an isobilateral leaf, the stomata are present almost in an equal number on both the surfaces. Mesophyll cells are not differentiated.

The secondary growth is quite distinct in most of the dicotyledons roots and stems. This helps in increasing the diameter of the roots and stems.

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EXERCISES

1. Define meristems.
2. Why is cambium considered to be a lateral meristem?
3. Name the type of plant tissue that has characteristically thin-walled cells and retains the capacity of division even at maturity.
4. What are sclereids?
5. Name the tissue which provides mechanical strength to the plant organs?
6. What is an annual ring?
7. The cross section of a plant material shows the following anatomical features under the microscope: (a) the vascular bundles are radially arranged (b) four xylem strands with exarch condition of the protoxylem. To which organ should it be assigned?
8. Name the tissue represented by the jute fibres used for making the ropes.
9. Indicate the location of cambium in a dicot stem.
10. Why a large number of stomata are present at the lower surface of the dicotyledonous leaves in the terrestrial plants?
11. Differentiate between:
    (a) closed and open vascular bundle
    (b) exarch and endarch condition of the protoxylem elements
    (c) heartwood and sapwood
    (d) metaxylem and protoxylem
    (e) metaphloem and protophloem
12. Mention two differences in the vascular bundles of sunflower and maize stems.
13. What is collenchyma? Explain its structure and function in plant body of a herbaceous angiosperm?
14. Describe the elements of phloem and xylem with the help of suitable diagrams.
15. What are the differences between the root hairs and stem hairs?
16. Describe the tissue system in plants.
17. Elaborate the process of secondary growth in a typical dicotyledonous stem/root with the help of diagrams.
18. Describe the internal structure of a dicotyledonous stem/root.
19. Draw and describe the internal structure of the leaf of a dicotyledonous plant. How would it differ from that of a monocot ?

CHAPTER 18

MORPHOLOGY OF ANIMALS

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Animals are multicellular organisms that take in preformed organic molecules in the form of food; they cannot synthesise these molecules from inorganic sources. Animals are heterotrophs and depend directly or indirectly for their nutrition on plants, photosynthetic algae or bacteria. Much of the diversity of animals evolved as they acquired the ability to capture and devour different types of food. In Chapter 6, you have come to know that animals are grouped into nonchordates and chordates. In this Chapter you will be acquainted with the salient features of earthworm and cockroach (nonchordates) as well as frog and rat (chordates). All of them are triploblastic, and bilaterally symmetrical and have attained organ system grade of organisation.

18.1 EARTHWORM

The earthworms are terrestrial animals and inhabit the moist soil. These are nocturnal animals and generally live in the upper layer of soil to a depth of about 30 to 45 cm. During daytime, they live in underground burrows; they make burrows partly by boring and partly by swallowing the soil. In the gardens, they can be traced by their faecal deposits known as **worm casting**. During rainy season, after a heavy rainfall, earthworms are seen in large numbers crawling on the ground. During winters, they drag leaves and vegetable debris into their burrows and plug the entrance to keep their burrows warm. Earthworm belongs to the Phylum Annelida, Genus *Pheretima* and Species *posthuma*. There are about 500 species of *Pheretima* and 13 of them are found in India.

External Morphology

The body of the earthworm is cylindrical, elongated and pointed anteriorly. The posterior end is slightly rounded. A fully-grown worm measures about 150 mm in length and 3 to 5 mm in width. The earthworm is reddish brown in colour and the dorsal surface is darker than the ventral surface. The ventral surface is distinguished by the presence of genital openings (pores). The dorsal surface of the body is marked by a dark median line (dorsal blood vessel) extending along the length of the body. The earthworm lacks a distinct head.

The entire body is made up of a series of distinct segments or metameres, which are separated from each other by intersegmental grooves (Fig. 18.1).

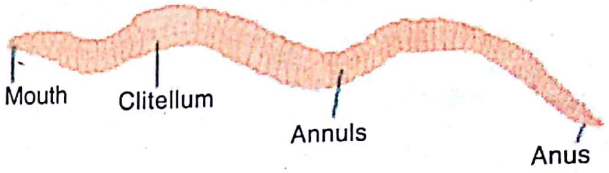

Fig. 18.1 *External morphology of earthworm*

The number of segments varies from 100 – 120. These segments are internally divided by septa. The four anterior segments in *Pheretima posthuma* show external segmentation but their corresponding internal septa are absent. The first segment at the anterior end of the body is called the 'buccal segment' or **peristomium**, which bears a very small terminal opening the mouth. A small projection is also present which hangs over the crescent shaped mouth and is called **prostomium.** The anus is located at the posterior end of the last segment. In a mature worm, a prominent circular band of glandular tissue surrounds segments from 14 to 16. These fused segments are designated as **clitellum**. Thus, the body is divisible into preclitellar, clitellar and postclitellar segments. Spermathecal apertures are four pairs and are

situated at the ventro-lateral sides of the intersegmental grooves between 5 and 6, 6 and 7, 7 and 8 and 8 and 9 segments. The single female genital pore is present in the mid-ventral line of the 14th segment. A pair of male genital pores is present on the ventro-lateral sides of the 18th segment. Numerous minute pores called nephridiopores, which are openings of the nephridia (excretory organs), open on the ventral surface of the body.

In all the body segments, except the first, last and clitellum, there is a ring of S-shaped **setae,** embedded in the epidermal pit at the middle of each segment (Fig.18.2).

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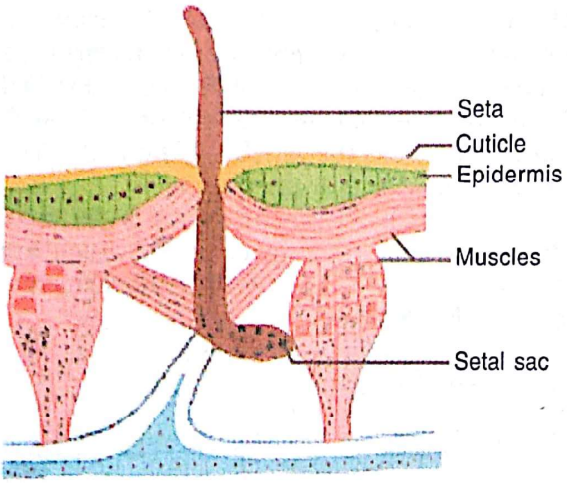

Fig. 18.2 *V.S. of body wall showing seta*

These are made up of chitin and help in locomotion by gripping the earth. These are not visible with naked eyes. Locomotion is performed by contraction and relaxation of the muscles and by setae for gripping on the soil. Also the intersegmental septa play an important role in locomotion.

Internal Morphology

The body wall of earthworm is covered externally by a thin - noncellular cuticle. The outermost cuticle is followed by an epidermis, two muscle layers and an innermost coelomic epithelium. The epidermis is made up of a single layer of columnar epithelial cells and contains many other types of cells, including the secretory gland cells. The muscle layers are composed of circular and longitudinal muscle fibres.

The excretory organs occur as segmentally arranged coiled tubules called nephridia (*sing.* Nephridium). These are of three types (Fig.18.3): (i) Those present on both the sides of intersegmental septa are called septal nephridia. These open into intestine. (ii) Those found attached to the lining of the body wall are called integumentary nephridia. These open on the body surface. (iii) Those found in the 4th, 5th and 6th segments in the form of three paired tufts are called pharyngeal nephridia. These nephridia exhibit a basic similarity in their structures.

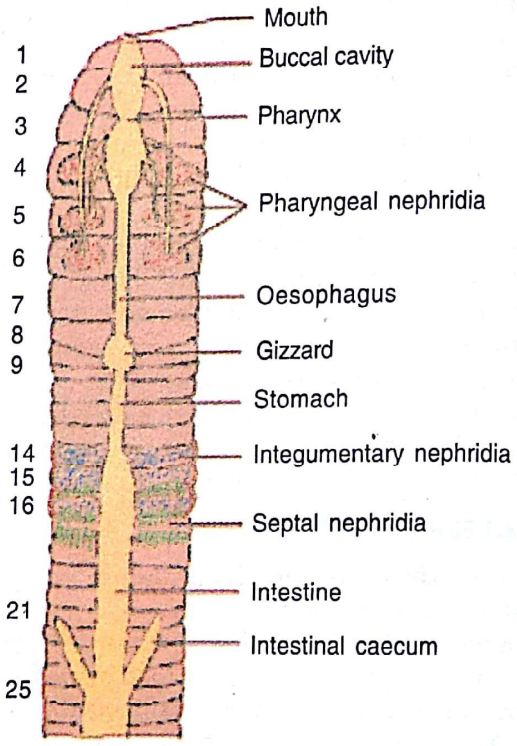

Fig. 18.3 *Arrangement of nephridia and alimentary canal*

The alimentary canal is a straight tube and runs from first to the last segment of the body (Fig.18.3). A terminal mouth (first segment) opens into the buccal cavity (1st–3rd segments) which leads to pharynx (4th segment). A small narrow tube, oesophagus (5th–7th segments), continues into a muscular gizzard (8th-9th segments). It helps in grinding the soil particles and decaying leaves, etc. The stomach extends from the 9th to 14th segments. The food of earthworm is decaying leaves and organic

matter (humus) mixed in soil. Calciferous glands are present in the stomach, which neutralise the humic acid. Extra calcium is passed out as calcite. Intestine starts from the 15th segment onwards and continues as a wide and straight tube till the last segment. A pair of short and conical intestinal caeca projects from the intestine on the 26th segment. The characteristic feature of the intestine is the presence of internal median fold of dorsal wall of intestine called **typhlosole** between the 26th to 95th segments. This enhances effective area of absorption after digestion. The alimentary canal opens to the exterior by a small rounded aperture called anus. Earthworm swallows soil rich in organic foodstuff formed as a result of decomposition of seeds, leaves, eggs and larvae of other animals. Insoluble and undigested food is given out along with soil through the anus as **worm casting**.

Pheretima represents a closed type of blood vascular system comprising blood vessels, capillaries, and heart with valves (Fig. 18.4). Blood glands, aggregates of follicles, are present on the 4th, 5th and 6th segments. These produce blood cells and haemoglobin. Haemoglobin remains dissolved in blood plasma. Blood cells do not possess haemoglobin. These are phagocytic in nature.

The nervous system is represented by ganglia arranged segment-wise on a ventral nerve cord. A ganglion is a mass of nerve cells. The nerve cord in the anterior region (3rd and 4th segments) bifurcates, laterally encircling the pharynx and uniting dorsally in the form of a **nerve ring**. The nerve ring with cerebral ganglia represents the brain. Sense organs are represented by epidermal, buccal and photoreceptors, and are poorly developed.

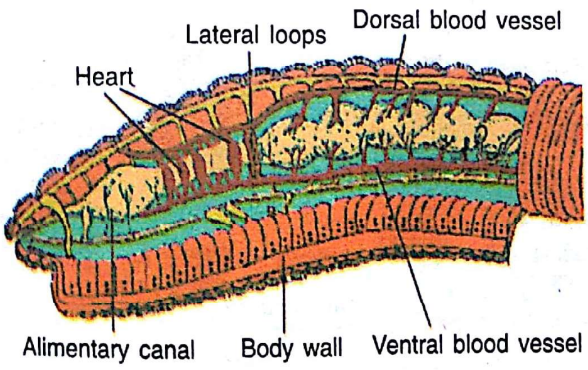

Fig. 18.4 *The major blood vessels and heart after removal of skin flap in the anterior region of earthworm*

Earthworm is hermaphrodite or bisexual i.e. testes and ovaries are present in the same individual (Fig. 18.5).

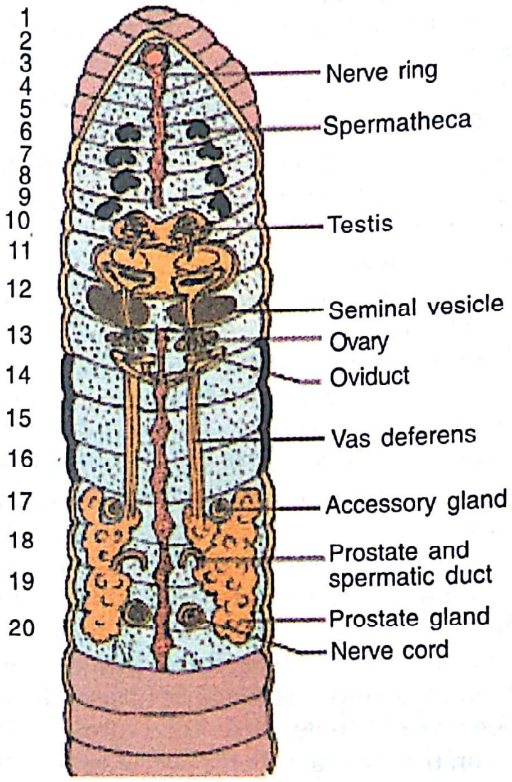

Fig. 18.5 *Reproductive system of earthworm*

There are two pairs of testes present in the 10th vasa and 11th segments. Their ducts, i.e. **vasa deferentia** run up to the 18th segment where they join the prostatic duct. Accessory glands are present on the ventral side of the 17th and 19th segments. The common prostate and spermatic duct opens to the exterior by a pair of male genital pores on the ventro-lateral side of the 18th segment. Four pairs of sac-like structures called spermathecae are found one in each of the 6th to 9th segments. They receive and store spermatozoa during copulation. One pair of ovaries is attached at the intersegmental septum of the 12th and 13th segments. Ovarian funnels are present beneath the ovaries which continue into oviduct, join together and open on the ventral side as a single median female genital pore on the 14th segment (Fig. 18.5). Development is direct, i.e. there is no larval forms.

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Interaction with Mankind

Earthworm is known as 'friend of farmers' because it makes small burrows and the soil becomes porous, facilitating respiration and penetration of developing plant roots. The decaying vegetation, which is taken by earthworm to burrows, enriches the soil. Soil particles are also broken to a fine state by the muscular gizzard. The digested and decayed vegetation and fine soil is deposited as worm casting. The nitrogenous wastes added to the soil are also used by plants. The process of increasing fertility of soil by earthworms is called **vermicomposting**. Earthworms are also used as food in some countries such as China, Japan, Myanmar (Burma), Australia, etc. Many tribal communities in India use earthworms in the form of medicine to cure bladder stones, jaundice, piles, diarrhoea, etc. Earthworm is also used as a bait for game fishing. Earthworms may cause some harm to man. They may damage young and tender plants. Besides, they also damage land by making burrows. This may cause seepage in areas where water table is high causing erosion of soil at slopes. The population of earthworm is declining due to uncontrolled pesticidal sprays and chemical fertilisers. Many animals such as frogs, birds and lizards, which prey upon them are also being affected indirectly resulting in ecological imbalance.

18.2 COCKROACH

Cockroaches prefer damp and warm places like sewage pipes, kitchen, bakeries, restaurants, warehouses, grocer's shop, etc. (Fig. 18.6). They are nocturnal and cursorial fast runner in habit. They possess three pairs of jointed appendages, and two pairs of wings which are characteristics of the class Insecta under the phylum arthropoda. They belong to the genus *Periplaneta*, which indicates their availability throughout the planet. The species available in India is known as *Periplaneta americana*. The word cockroach is derived from the Spanish word *cucaracha*. It is believed to be a native of America (Mexico) and has spread with man throughout the planet. About 2600 species of cockroach are known from all over the world.

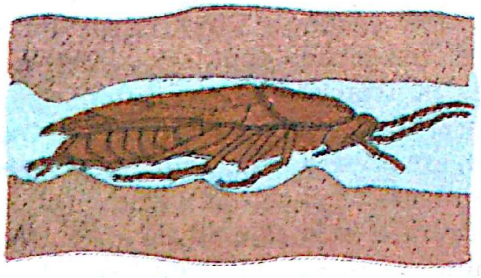

Fig. 18.6 *Cockroach in hiding place*

External Morphology

The body of cockroach is elongated and dorsoventrally flattened and reddish-brown in colour. Males usually measure 35-40 mm in length and about 10-12 mm in width but the females are slightly smaller. The entire body is covered by a hard and brown chitinous exoskeleton. In each segment exoskeleton has hardened plates called sclerites that are joined to each other by thin and flexible articular membranes.

The body is segmented and divisible into three distinct regions – head, thorax and abdomen (Fig. 18.7). The **head** is triangular and lies anteriorly at right angles to the longitudinal body axis. The head is formed by the fusion of six segments and is highly mobile in all directions due to a flexible neck. The exoskeletal covering of the head is called head capsule. The head bears

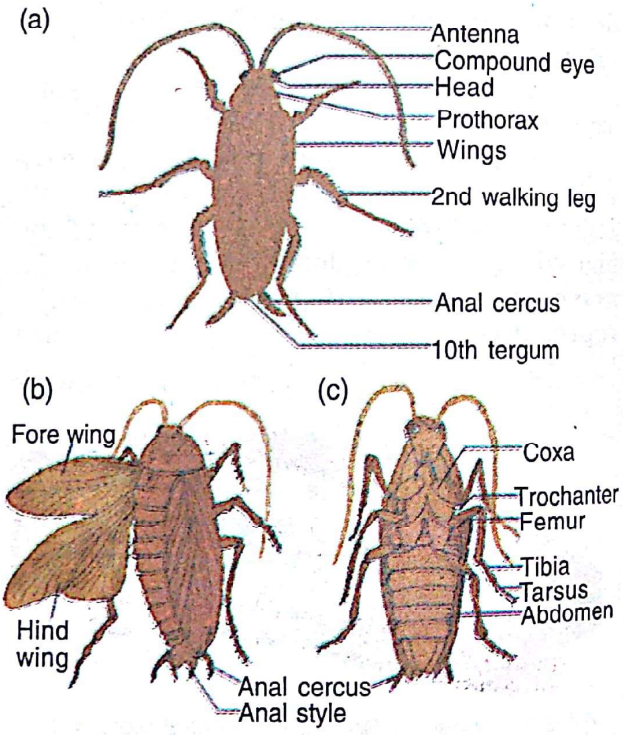

Fig. 18.7 *External morphology of cockroach showing (a) dorsal view of female (b) dorsal view of male after stretching of wings (c) ventral view of female*

a pair of sessile (without stalk) compound eyes, one on each of its sides. A pair of thread-like long antennae (sing. antenna) arise from membranous sockets lying in front of the eyes; they can be moved in all directions and are very sensitive. Anterior end of the head bears the mouth which is provided with appendages, collectively called mouth parts, which are used in chewing, cutting and swallowing. The mouthparts consist of a pair of mandibles and maxillae, **labium** forming lower lip and a **labrum** forming the upper lip (Fig. 18.8). Within the cavity enclosed by mouthparts there is a median flexible lobe called **hypopharynx**, which acts like a tongue.

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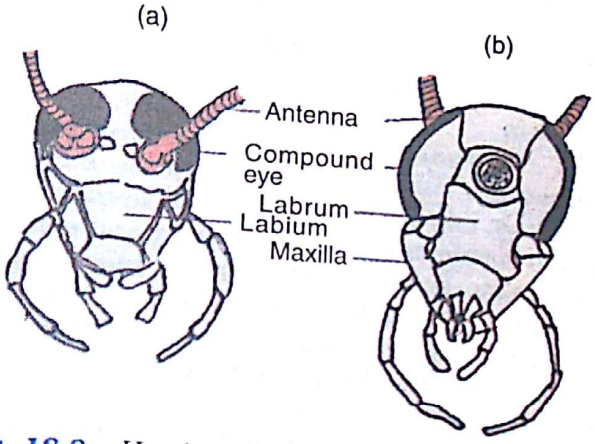

Fig. 18.8 *Head region of cockroach showing mouth parts (a) front view (b) back view*

The **thorax** consists of 3 segments – the prothorax, mesothorax and metathorax. The head is connected with the thorax by a short extension of the prothorax, the neck. A large sclerite covers the prothorax and shields the mesothorax. The mesothorax and metathorax have independent coverings called sclerite. Each thoracic segment bears a pair of walking legs. Each walking leg consists of five segments: (a) a short and broad **coxa**, (b) a triangular short and rod-like **trochanter**, articulated with (c) a long spout, spiny **femur**, (d) a spring **tibia** which represents the largest segment, and (e) a long **tarsus**. The first pair of wings arises from mesothorax and the second pair from metathorax. The fore-wings **(mesothoracic)**, known as **tegmina,** are opaque dark, leathery in texture and used to cover the hind wings **(metathoracic)**, when at rest. The hind wings are transparent, membranous, thickened by veins and are used in flight.

The abdomen in both males and females consists of 10 segments. In females, sclerites of the 8th and 9th segments are overlapped by corresponding sclerites of the 7th segment. The seventh sternum is boat-shaped and together with eighth and ninth sterna forms a **brood** or **genital** pouch. In males, only 8th segment is overlapped by the 7th segment.The tenth segment bears a pair of jointed filamentous structures called anal cerci (*sing.cercus*). On ventral side of male, the 9th segment bears a pair of short, thread-like anal styles which are absent in females.

Internal Morphology

The body remains covered by a cuticle, which is impermeable to water. Numerous fine tubules originating from lower epidermis traverse the cuticle. The alimentary canal is present in the body cavity. It is divided into three regions: fore-gut, mid-gut and hind- gut. The mouth opens into a short tubular pharynx, which passes and bends into a narrow, tubular passage called oesophagus. Oesophagus opens into a sac-like structure, crop (Fig 18.9). The crop is followed by a **gizzard** or **proventriculus**. It has an outer thick layer of circular muscles and a thick, inner cuticle forming six highly chitinous plates called **teeth.** Gizzard is designed in such a manner

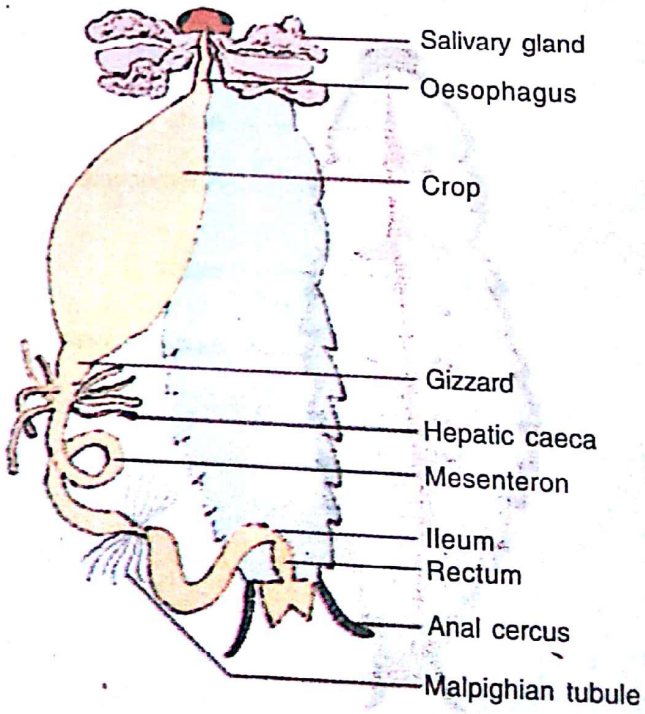

Fig. 18.9 *Alimentary canal of cockroach*

that it helps in grinding the hard food particles. The entire foregut is lined by cuticle. The mid-gut or mesenteron is a narrow tube of uniform diameter without inner lining of cuticle. A ring of blind caecae (eight in number) called **hepatic caecae**, which secrete digestive juice, is present at the junction of fore and mid-gut. At the junction of mid-gut and hind-gut there is a ring of about 150 thin yellowish **Malpighian tubules**, which help in excretion. The hind-gut is slightly broader than the mid- gut and is lined internally by cuticle. It opens by an anus, lying posteriorly below the 10th tergum. The digestive juices are produced not only by the wall of mesenteron and hepatic caeca but also by a pair of large salivary glands which open into the pharynx.

The blood vascular system of cockroach is of open type. It means the blood vessels are poorly developed and open into spaces rather than the capillaries. All the visceral organs are bathed in blood (also called haemolymph). Haemolymph is composed of colourless plasma and corpuscles called haemocytes. The respiratory pigments are absent in the blood. To regulate blood flow in the haemocoel, an elongated tube with muscular wall is lying mid dorsally in the thorax and abdomen consisting of 13 funnel-shaped chambers, which are arranged segmentally (Fig.18.10). Each chamber represents one **heart**. At the lateral sides of each chamber two pores, ostia (sing. ostium), are present one on each side. Ostium is guided by a valve to allow blood flow only in one direction i.e. from haemocoel to the inner chamber of the heart.

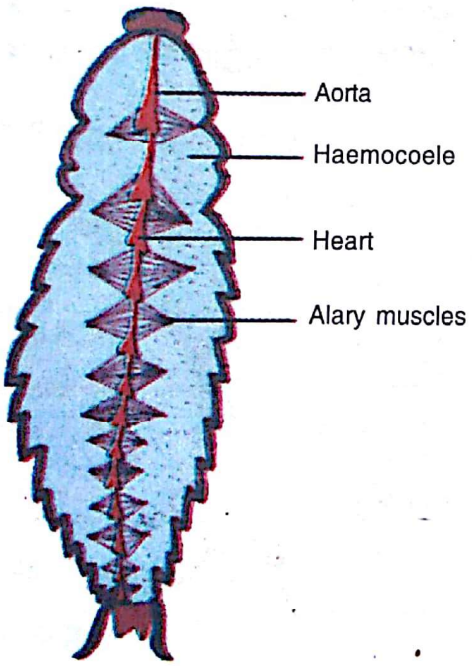

Fig. 18.10 *Heart of cockroach (dorsal view)*

The respiratory system consists of a network of trachea, the opening of which is called spiracle. Air enters through spiracles, which are ten pairs in number; and are arranged segmentally on lateral sides. Two pairs lie in the meso and metathorax and eight pairs in the abdomen. The opening of the spiracles is regulated by sphincters. The tracheal tubes are sub-divided into tracheoles, which branch within the tissues. Air reaches upto tracheoles, and the body fluid in the tracheoles exchanges dissolved oxygen and carbon dioxide with the cells. The exchange of gases takes place by diffusion.

In cockroach excretion is performed by Malpighian tubules. Each tubule is lined by glandular, ciliated cells with brush-border. They absorb nitrogenous waste product and convert it into uric acid through various biochemical processes. The uric acid is excreted through hind-gut. Hence this insect is called **Uricotelic**. In addition, fat bodies, nephrocytes, cuticle and urecose glands also help in excretion.

The main nervous system consists of a series of fused, segmentally arranged ganglia joined by paired longitudinal connectives on the ventral side. Three ganglia lie in thorax and six in the abdomen. In the head region the brain is represented by supra-oesophageal ganglion which supplies nerves to antenna and compound eyes. It joins sub-oesophageal ganglion by two circum-oesophageal commissures. These constitute a characteristic nerve ring around the oesophagus. In cockroach **sense organs** are antenna, eyes, maxillary palps, labial palps and anal circi, etc. The antennae, palps and circi are very sensitive tactile sense organs, which help in detection of food and various objects. The compound eyes are situated at the dorsal surface of the head in the form of black kidney-shaped patches. Each eye consists of about 2000 hexagonal facets or ommatidia (sing. ommatidium), which are covered externally by transparent cuticle called cornea. Ommatidium is composed of corneal lens, a pair of corneagen cells, refractive crystalline cone distal pigment cells around the

pigment cone and retinula forming rhabdome, basal pigment cell, basement membrane and optic nerve. With the help of several ommatidia a cockroach can receive several images of an object. This kind of vision is known as **mosaic vision**. This is how the compound eye of the cockroach is adapted for detecting movement more efficiently than an eye of a vertebrate.

Male reproductive organs comprise a pair of small trilobed testes lying one on each lateral side in the 4th –5th abdominal segments. From each testis arises a thin tube called **vas deferens**, which opens into ejaculatary duct through the **seminal vesicle**. The ejaculatory duct opens into male gonopore situated ventral to anus. A characteristic mushroom-shaped gland is present in the 6th –7th abdominal segments, which functions as an accessory reproductive gland. The external genitalia are represented by chitinous, asymmetrical structures, surrounding the male gonopore, at the end of the abdomen, called male gonapophysis or **phallomere** (Fig.18.11). The sperms are stored in seminal vesicles and are glued together in the form of bundles called **spermatophores**, which are discharged during copulation. Female organs comprise two large ovaries lying laterally in the 4th, 5th and 6th segments. Each ovary is formed of a group of 8 ovarian tubules or ovarioles, containing a chain of developing ova. Oviducts of each ovary unite into a single, median oviduct, which is also called **vagina**, which opens into the genital chamber. The female accessory glands are a pair of branched, collateral glands, which open into the dorsal side of the genital chamber. A pair of spermatheca is present in the 6th segment, which opens into the genital chamber on a small papillae (Fig.18.11).

Interaction with Mankind

Cockroach causes damage to the household material including clothes, shoes, purses, etc. It also eats and destroys foodstuffs. The food contaminated by it gives typical smell, making it unpalatable. While living in sewage pipes and gutter holes, these also carry harmful germs of

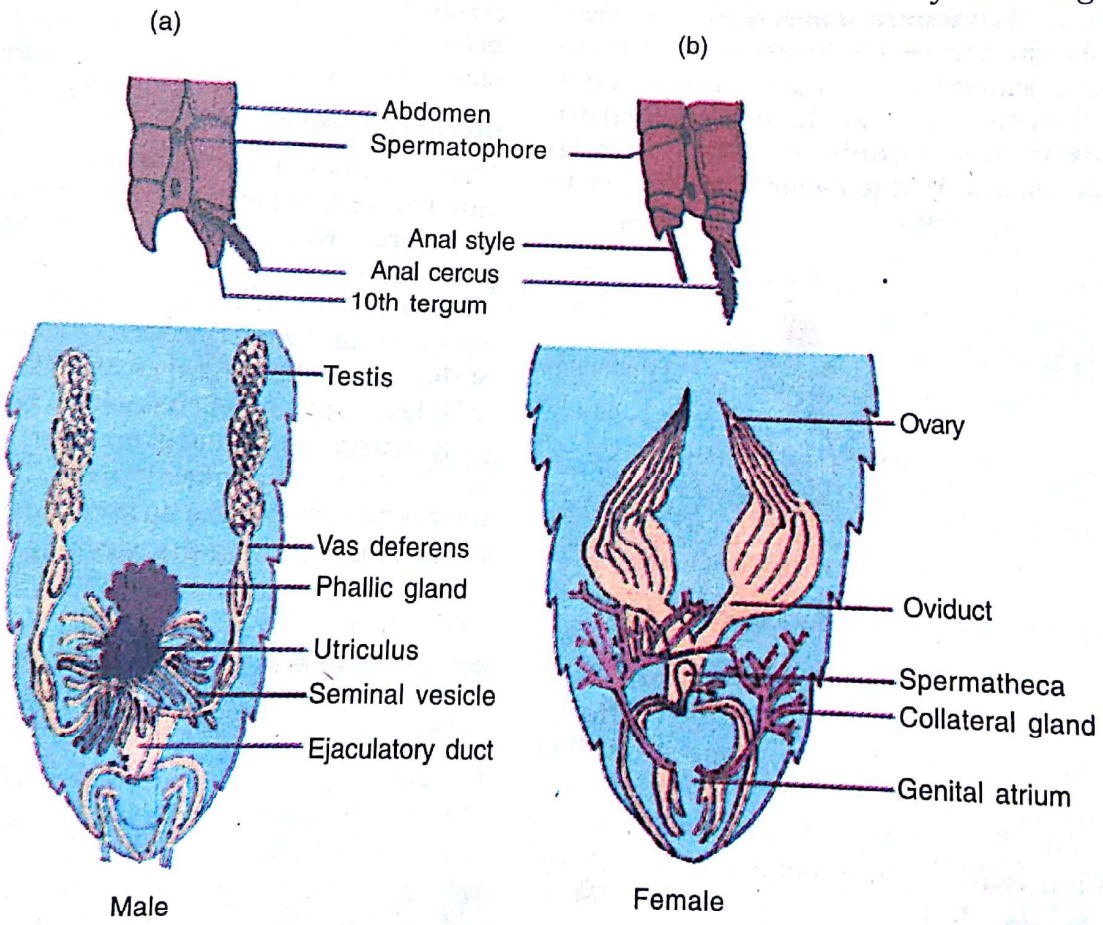

Fig. 18.11 *Reproductive organs of cockroach (a) male (b) female*

diseases like cholera, diarrhoea, tuberculosis, typhoid, etc. In some South American countries and in Myanmar people eat cockroach. Many amphibians, birds, lizards and rodents prey upon cockroach. Thus, it serves as a part of food chain. It is also used as a safe experimental animal for laboratory exercises and biological research work since it can be obtained easily and without causing much disturbance to the ecosystem.

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18.3 FROG

The most common frog found in India is the Indian bullfrog. It is the largest frog and is named so because of its large size and loud call. Indian bullfrog is found in freshwater marshes, ditches, ponds, and shallow lakes. They are also reported from running waters, and are seen in large numbers during rainy season as they breed during this period. They undergo **aestivation** (summer sleep) in summer and **hibernation** (winter sleep) in winter. During scorching summer they protect themselves by burrowing in damp and soft soil. They rest in a squatting posture, i.e. hind-limbs folded inside and fore-limbs upright (Fig.18.12). They are carnivorous (feeding upon other animals, insects, etc.), poikilotherms, i.e., the body temperature changes with environment. They develop protective colouration to camouflage, that is, to hide in surroundings.

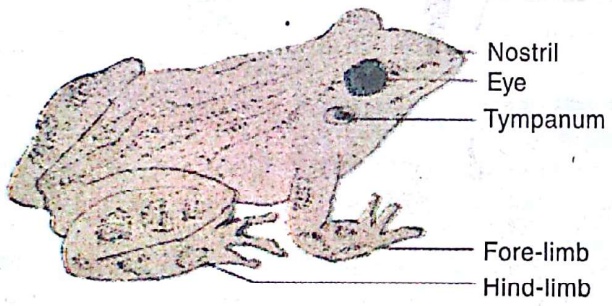

Fig. 18.12 *External morphology of frog*

Frogs belong to the Phylum Chordata, Subphylum Vertebrata or Craniata, Superclass Gnathostomata, Class Amphibia and Genus *Rana*. The most common species is known as *Rana tigrina*. The scientific name of common toad is *Bufo melanostictus*. Frogs exhibit sexual dimorphism. Male and female are distinguishable externally only during breeding season when the males develop nuptial pad in the thumb. Vocal sac in males produces louder sound as compared to the females, which are devoid of vocal sac.

External Morphology

The body of frog is adapted for swimming in water, burrowing into mud and leaping on land. It is streamlined with soft, moist and slimy skin. Skin is made up of epidermis and dermis. Mucous glands are present in the dermis and their ducts open at the surface. Blood capillaries and pigment cells (chromatophores) are present in the dermis. Skin is without scales or any other cover or exoskeleton. The mature animal is 18 – 20 cm in length and 5-8 cm. in width. Colour of dorsal skin of frog is generally olive green with dark irregular lines and spots. On ventral side, it is uniformly pale yellow. Body is divisible into head and trunk, neck is absent. The trunk is provided with a pair of fore and hind-limbs. The hind-limbs are much larger and muscular than the fore-limbs. Fore limbs end in four digits and the hind limbs end in five digits. Eyes are provided with nictitating membrane for protection and positioned dorso-laterally on the head. A little below the eye a tympanum is present on either side of head that receives sound signals.

Internal Morphology

Since the frogs are carnivorous, their alimentary canal is short in length. The mouth is present as a terminal, wide opening. It opens into bucco-pharyngeal cavity, which contains numerous maxillary teeth arranged along the margin of the upper jaw and vomerine teeth present in the roof of the bucco-pharyngeal cavity. The lower jaw is toothless. Opening of eustachian tube, vocal sac (only in male) gullet and glottis can be seen clearly in the bucco-pharyngeal cavity. The muscular tongue is bilobed at the tip and free from behind.It is used for capturing the prey (Fig. 18.13).

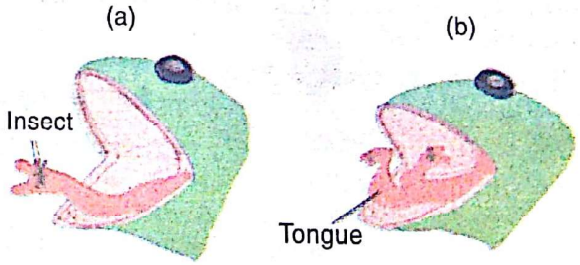

Fig. 18.13 *Capturing of prey by frog. (a) forward stroke of tongue (b) backward stroke and infolding of tongue to ingest the prey*

The gullet opens in narrow and short tube-like oesophagus, which continues in large and distended stomach. It contains a thick muscular layer, which helps in converting food into chyme. It secretes gastric juice containing HCl and proteolytic enzymes. Stomach is followed by a coiled small intestine. Intestinal wall has numerous finger-like folds, called **villi** and **microvilli,** projecting into its lumen to enhance surface area of absorption for digested food. The first part of the small intestine lying parallel to stomach is called **duodenum**. Intestine continues into a wider rectum, opening into cloaca. The urinary bladder opens into cloacal chamber through the ureter (Fig.18.14).

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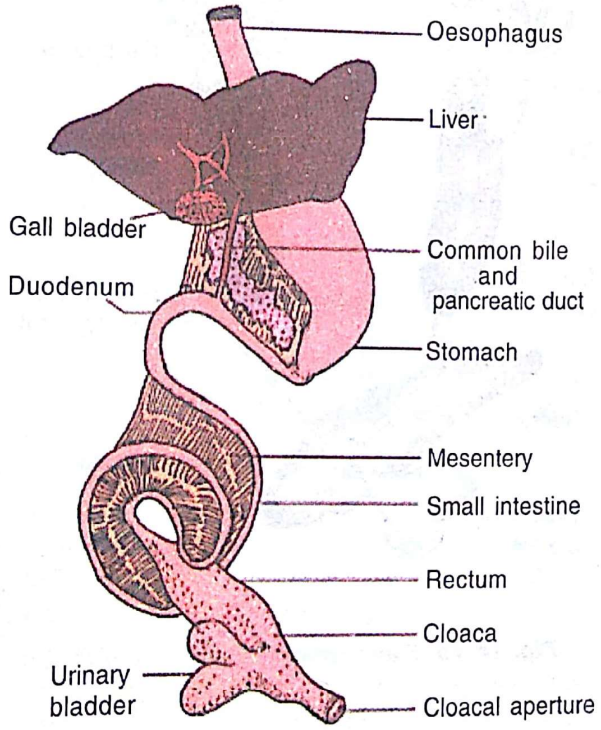

Fig. 18.14 *Alimentary canal and digestive glands of frog*

The gastric and intestinal glands occur in the walls of stomach and intestine, respectively. The other important digestive glands associated with the alimentary canal are **liver** and **pancreas**. Liver secretes bile, which is temporarily stored in gall bladder before being released into the duodenum. Bile helps in digestion of food by changing its pH from acidic to alkaline and by emulsifying the fat. Liver does not secrete any digestive enzyme. Pancreas is an irregular, elongated gland, situated in a thin mesentery and lies parallel to the stomach. It produces pancreatic juice containing digestive enzymes like trypsin, amylopsin, etc.

Frogs respire both on land as well as in water. Skin acts as an aquatic respiratory organ. Exchange of gases takes place by diffusion. Buccal cavity and lungs act as aerial respiratory organs. The respiration by lungs is called pulmonary respiration. The lungs are a pair of elongated, pear-shaped, pink coloured sac-like structures present in thoracic (upper) part of the trunk region. During aestivation and hibernation, they respire by skin (cutaneous respiration).

The circulatory system of frog consists of blood vascular system and lymphatic system. It is of closed type and represents single circulation, which means that both the oxygenated and the deoxygenated blood enters the heart and get mixed in the ventricle. The blood vascular system comprises heart, blood vessels and blood, whereas the lymphatic system consists of lymph, lymph channels and lymph nodes. Both systems are inter-connected because lymph channels open into large venous channels. **Heart** is a three chambered, muscular structure situated in the upper part of the body cavity. A thin membrane called pericardium covers it. It has two upper chambers (auricles) which open separately into a single lower chamber, ventricle. A short sac-like conus arteriosus is present on the ventral side of the heart over the larger right auricle. On dorsal side, a triangular structure, called sinus venosus is present in which all the three main vena cava (right and left anterior and posterior) are open. A definitely arranged network of arteries and veins form the arterial and venous systems. Frogs also possess two well developed portal systems: renal portal and hepatic portal systems. In these the concerning veins originate from intestine and break up into fine capillaries in the respective organs, kidney (renal portal system) or liver (hepatic portal system). Three types of **blood corpuscles** are present in plasma, viz. RBC (red blood corpuscles) or erythrocytes, WBC (white blood corpuscles) or leucocytes and thrombocytes. RBC's contain red coloured respiratory pigment namely **haemoglobin**. RBCs are nucleated. The **lymph** is a filtered out fluid from blood capillaries. It is different from blood, lacking a few proteins and RBCs.

The nervous system is organised into a central nervous system (brain and spinal cord), a peripheral nervous system (cranial and spinal nerves) and an autonomic nervous system (sympathetic and parasympathetic chains of ganglia). There are ten pairs of cranial nerves. Brain is enclosed in a bony structure or brain box (cranium), which has two occipital condyles for attachment with the first vertebra (atlas). The brain is divided into fore-brain, mid-brain and hind-brain. Forebrain includes olfactory lobes, paired cerebral hemispheres and unpaired diencephalon. The mid-brain is characterised by a pair of optic lobes. Hind-brain consists of cerebellum and medulla oblongata. The medulla oblongata passes out through the foramen magnum and continues into **spinal cord**, which is contained in the vertebral column.

Frog has five types of **sense organs**, namely organs of touch (sensory papillae), taste (taste buds), smell (nasal epithelium), vision (eyes) and hearing (tympanum and internal ears). Out of these, eyes and internal ears are well-organised structures and the rest are cellular aggregations around nerve endings. **Eyes** in a frog are a pair of spherical structures situated in the orbit. These are simple eyes (possessing only one unit). The wall of eye is composed of three layers, sclerotic, choroid and retina. The cornea is transparent. **External ear** is absent in frogs and only tympanum can be seen externally. **Middle ear**, named as tympanic cavity, is filled with air. It passes vibrations to **internal ear** or membranous labyrinth. It is present in a fluid-filled otic or **auditory capsule**. It has two sac-like bodies (utriculus and sacculus) and three semicircular canals. The ear is an organ of hearing as well as balancing (equilibrium).

The main organ of excretion is a pair of kidneys. These are compact, dark red and bean like structures situated little posteriorly in the body cavity on both sides of vertebral column. The frog excretes urea thus, is a ureotelic animal. It is carried by blood into the kidney where it is separated and excreted. Each kidney is composed of several structural and functional units called **uriniferous tubules** or **nephrons**. Ureters emerge from the kidney as urinogenital ducts in the male, which opens into the cloaca. A thin-walled urinary bladder is present ventral to the rectum, which also opens in the cloaca.

The chemical coordination of various organs of the body is manifested by hormones, which are secretions of the endocrine glands. The prominent endocrine glands found in a frog are pituitary, thyroid, parathyroid, thymus, pineal body, pancreatic islets, adrenals and gonads.

Male reproductive organs consist of a pair of yellowish ovoid testes, which are found adhered to the upper part of kidneys by a double fold of peritoneum called mesorchium. **Vasa efferentia** are 10-12 in number and after arising from testes run through the mesorchium and enter the kidneys of their side. In kidneys, these open

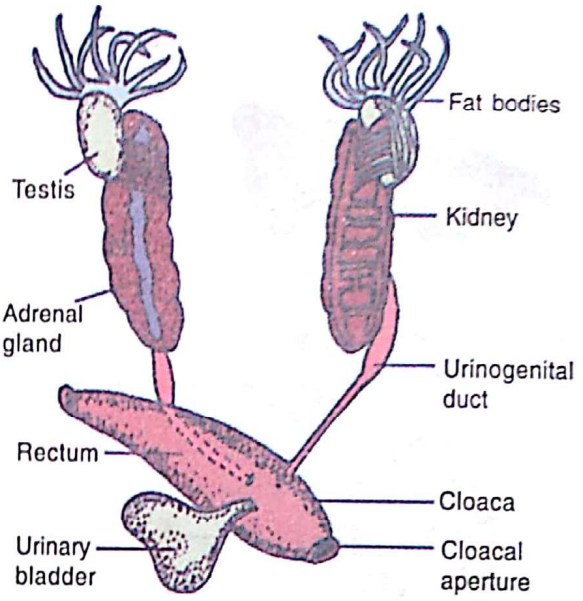

Fig. 18.15 *Male reproductive organs of frog*

into **Bidder's canal**, which finally communicates with the urinogenital duct, which comes out of the kidneys and finally opens into the **cloaca**. The cloaca is a small, median chamber that is used to pass faecal matter, urine and sperms to the exterior (Fig.18.15).

A pair of ovaries, situated near kidneys, comprises the female reproductive organs. However, these have no functional connection with kidneys. A pair of oviduct opens into the cloaca, separately. Ovaries produce ova by oogenesis. A mature female can lay 2500 to 3000 ova at a time (Fig.18.16). Fertilisation is external. Development is indirect involving a larval stage called tadpole.

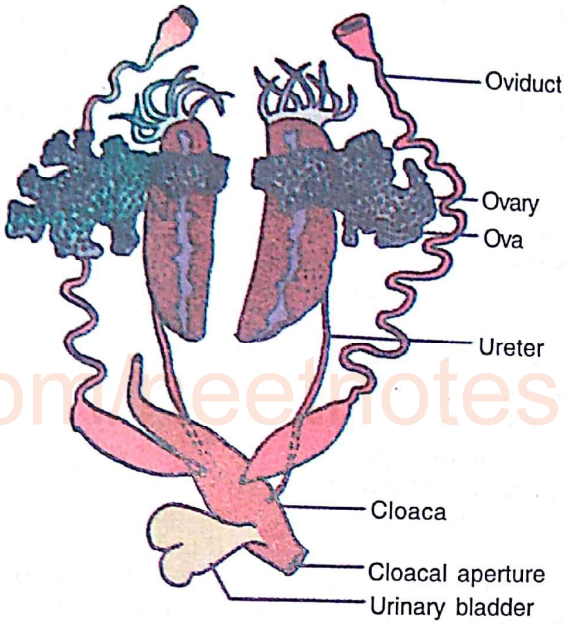

Fig. 18.16 Female reproductive organs of frog

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Interaction with Mankind

Frog is a beneficial animal for human-being. Frog eats up insects and thus protects the crop. This saves expenditure on insecticides. Frog has been used as an experimental material for teaching and research. Due to habitat loss and large scale use of this animal for dissections it has become a threatened species. Therefore, use of frog for classroom exercise should be discouraged as far as possible. The muscular legs of frog are used as food by man in some parts of India and many other countries. These were exported in huge quantities, but it is legally banned now. Small froglets are used as fish bait. Frogs maintain ecological balance because these serve as an important food link in ecosystem. Due to over use of pesticides, insecticides and other agro-chemicals they are being vanished at a fast rate causing loss to biodiversity. This is creating environmental problems.

18.4 RAT

Rat inhabits holes and burrows in cultivated fields. It makes its own burrows and feeds on stored grains. By nature it is nocturnal but in human habitations it remains active during day hours also. Rat is a member of the Phylum Chordata, Subphylum Vertebrata or Craniata, Superclass Gnathostomata and Class Mammalia. The genus of rat is *Rattus*. There are about 137 species of the genus *Rattus* of which the most common species are: *R. rattus* (black rat) and *R. norvegicus* (common brown rat).

External Morphology

The size of the adult rat is about 20 cm long from nostril to tail tip. Its body is elongated and covered with hair, except nose, lips, palm and soles, which are brown black. The rat is characterised by having a fusiform body consisting of a tapered head, neck, a slender trunk and a long tail, which is covered by vestigial scales and very few remnant hairs. It acts as a balancing organ. Head is broader posteriorly and tapers anteriorly as snout. A pair of nostrils is present above the mouth opening, which leads into nasal passage. Eyes are large, paired and located on the lateral sides of the head. A movable upper eyelid and a fixed lower eyelid guard eyes, which are provided with eyelashes. The nictitating membrane is reduced. The head bears a pair of external ear or pinna at its postero-lateral position. The mouth is sub-terminal and located beneath the nostrils. It remains guarded by upper and lower lips. The upper lip is usually with a cleft in the centre made by a vertical groove. The teeth are heterodont (different types) and thecodont, i.e. present in sockets or gums. There are two incisors and six molars in each jaw. The incisors grow throughout life and act as cutting teeth. Due to the absence of enamel on the surface a sharp cutting edge is maintained. The canines and premolars are absent. On both sides of nostrils, the bristles are developed as specialised tactile hairs and are known as *pili tactiles* or vibrissae (Fig. 18.17).

The trunk bears two pairs of limbs, two fore limbs and two hind limbs. Forelimbs are smaller than the hindlimbs. Each limb is made up of proximal segment (stylopodium), middle segment (zeugopodium) and distal segment (autopodium). Five digits are present in autopodium of each limb. The first digit is thumb or polex, which is much reduced on the forelimb and has a flattened nail unlike the more rounded nails of the other digits. Nail is a keratinized structure occupying position above the distal phalanx of each digit. Typical walking pads (tori) are

present in the palm. The forelimb has five apical pads, three inter-digital pads and two basal pads.

The anus is placed at the posterior terminal end. In the females, the urinary and genital apertures are located beneath the anus and open separately into vulva. In the males, the urinary and genital apertures open commonly through the urethra into a muscular penis. In females, six pairs of nipples are present on the ventral surface of the trunk.

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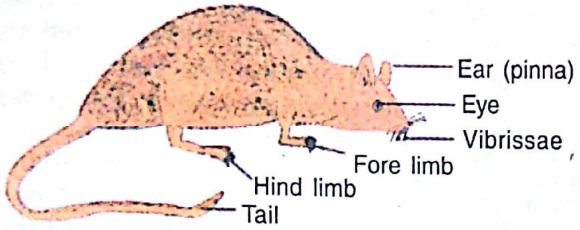

Fig. 18.17 *External morphology of rat*

Internal Morphology

The integument is made up of multilayered epidermis and a dermis. The lowermost layer of epidermis is highly proliferative and called **stratum germinativum** and the outermost one is the **stratum corneum**. Blood vessels are present in the dermis. Hair and glands are found in the skin. The major skin glands are sweat glands, sebaceous glands, mammary glands (modified sweat glands), lacrimal or tear glands, meibomian glands, wax glands, etc. (Fig.18.18).

The mouth opens in the buccal cavity that is surrounded by the vestibule, which is a space between the lips, cheeks and teeth. Pharynx is a common chamber for the passage of food and air. Oesophagus is a short tube situated dorsal to the trachea and it leads into the stomach. Stomach leads into small intestine, which is divided into three parts duodenum, jejunum and ileum. Ileum opens into caecum, which in turn, opens into the first part of large intestine, the colon. The colon leads into rectum, which opens outside through the anus (Fig.18.19).

Four pairs of salivary glands for secretion of saliva are found in the buccal cavity. These are parotid, mandibular, sublingual and infra-orbital glands. The saliva contains the enzyme ptyalin that helps to digest starch of the food. The liver is the largest digestive gland and is located below the diaphragm. It has four lobes and its cells (hepatocytes) secrete bile that helps in digestion. Gall bladder is absent. Pancreas is very diffused and present between the duodenal loops and secretes pancreatic juice, containing enzymes such as trypsinogen, amylopsin and lipase.

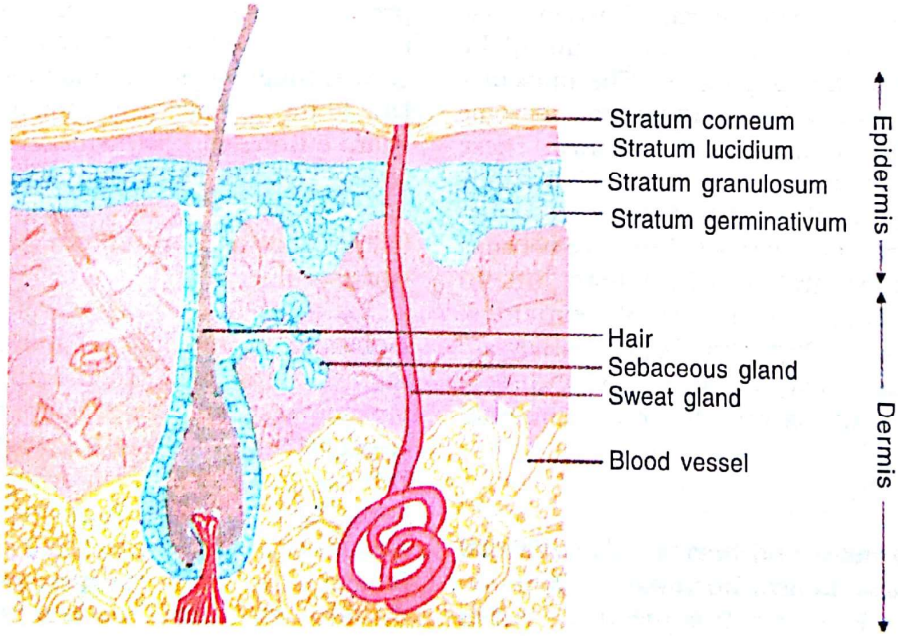

Fig. 18.18 *V.S. of skin of rat*

SUMMARY

Earthworm, Cockroach, Frog and Rat show characteristic features in body organisation, segmentation and symmetry, etc. In *Pheretima posthuma* (earthworm) the body is metamerically segmented. It is a terrestrial animal and lives in burrows. The faecal material deposits called worm castings, help in tracing this animal. A dark median line is present on the dorsal surface. It is reddish brown in colour. The body is covered by cuticle. All segments of its body are alike except the 14th, 15th and 16th segments, which are thick and dark and glandular, forming clitellum. A ring of S-shaped chitinous setae is found in each segment. These setae help in locomotion. On the ventral side spermathecal openings are present in between the grooves of 5 and 6, 6 and 7, 7 and 8 and 8 and 9 segments. Female genital pores are present on 14th segment and male genital pores on 18th segment. Excretory organs are nephridia. These are of three types - septal, integumentary and pharyngeal. The alimentary canal is a narrow tube made up of mouth, buccal cavity, pharynx, gizzard, stomach, intestine and anus. The blood vascular system is closed type with heart and valves. Nervous system is represented by ventral nerve cord. Earthworm is hermaphrodite. Two pairs of testes occur on the 10th and 11th segment, respectively. A pair of ovaries are present on 12 and 13th intersegmental septum. It is a protoandrous animal with cross-fertilization. Fertilization and development of new ones take place in cocoon secreted by the glands of Clitellum.

Cockroach (*Periplaneta americana*) is found all over the world. Its body is covered by chitinous exoskeleton. It is divided into head, thorax and abdomen. Segments bear jointed appendages. There are three segments of thorax, each bearing a pair of walking legs. Two pairs of wings are present one each on 2nd and 3rd segment. There are ten segments in abdomen. Alimentary canal is well developed with a mouth surrounded by mouth parts, a pharynx, oesophagus, crop, gizzard, fore and midgut, hindgut and anus. Hepatic caecae are present at the junction of fore and midgut. Malpighian tubules are present at the junction of midgut and hindgut and help in excretion. A pair of salivary gland is present near crop. The blood vascular system is open type. Respiration takes place by network of tracheae. Trachea opens outside with spiracles. Nervous system is represented by segmentally arranged ganglia and ventral nerve cord. A pair of testes is present in 4th and 5th segments and ovaries in 4th, 5th and 6th segments. Fertilization is internal. Female produces 10-40 ootheca bearing developing embryos. After rupturing of single ootheca sixteen young ones, called nymphs come out.

The Indian bullfrog, *Rana tigerina*, is the largest frog found in India. Body is covered by skin. There are no derivatives found in the skin. Mucous glands are present in the skin which is highly vascularised and helps in respiration. Body is divisible into head and trunk. Maxillary teeth are present along the margin of upper jaw. These are absent in the lower jaw. A muscular tongue is present, which is bilobed at the tip and is used in capturing the prey. The alimentary canal consists of oesophagous, stomach, intestine and rectum, which open into the cloaca. The main digestive glands are liver and pancreas. It can respire in water through skin and through lungs on land. Circulatory system is closed with single circulation. The blood circulatory system is connected to lymphatic system. RBCs are nucleated. Nervous system is organised into central, peripheral and autonomic. The organs of urinogenital system are kidneys and urinogenital ducts, which open into the cloaca. The male reproductive organ is a pair of testes. The female reproductive organ is a pair of ovaries. A female lays 2500-3000 ova at a time. The fertilization and development are external. The eggs hatch into tadpoles, which metamorphose into frogs. Tadpoles show regeneration of limb and tail.

Common Indian black rat (*Rattus rattus*) is nocturnal, inhabiting holes and burrows. Rat is about 20 cm in length, its body is covered by hair, except at few places and is characterised by a fusiform body. Integument is made up of epidermis, dermis and their derivatives. Digestive system consists of buccal cavity, pharynx, oesophagus, stomach, small intestine, large intestine and associated glands. The gall bladder is absent and

pancreas is diffused in rat. Closed and double circulatory system is present in rats. The heart is four chambered, located in mid line and is surrounded by pericardium. Arterial and venous system, respiratory, excretory, endocrine and nervous systems are well developed. Male reproductive system consists of a pair of testis, epididymis, vas deferens, urethra, penis, spermatic cord and five accessory glands. Female reproductive organs consist of a pair of ovary, fallopian tube, uterus, clitoris, vagina and three associated glands. Six to eight young ones are produced in a litter.

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EXERCISES

1. Answer in one word or one line.
   (i) Give the scientific name of earthworm.
   (ii) . Give the common name of *Periplanata americana*.
   (iii) How many spermatheca are found in earthworm?
   (iv) What is the position of ovaries in cockroach?
   (v) What is the name of tracheal opening in cockroach?
   (vi) How many segments are present in the abdomen of cockroach?
   (vii) Name the part of alimentary canal of frog where the gullet opens.
   (viii) What is the function of vibrissae?
   (ix) Where do you find Malpighian tubules?
2. Answer the following:
   (i) Give systematic position of a rat.
   (ii) What is the function of nephridia?
   (iii) Give three differences between frogs and toads.
   (iv) Name the parts of the alimentary canal of a rat.
   (v) What do you understand by open type of circulatory system? Give a suitable example.
   (vi) How many types of nephridia are found in earthworm based on their location?
   (vii) Give the names of male reproductive organs of a rat.
3. Draw a labelled diagram of the reproductive organs of an earthworm.
4. Write the names of appendages associated with the head of a cockroach.
5. Draw a labelled diagram of alimentary canal of a cockroach.
6. Give a note on organs of excretion in a frog.
7. Describe the female reproductive organs of a rat and compare thses with that of a frog.
8. Describe the alimentary canal of an earthworm and its interactions with mankind.

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CHAPTER 19

ANIMAL TISSUES

As you know by now, all the life activities of the body such as nutrition, respiration, excretion, etc. are performed by the single cell of unicellular organisms (protozoans). Origin and evolution of multicellularity resulted in the attainment of larger size of body with complexity in organisation. In order to perform various life activities in such multicellular plants and animals (metazoans), there developed a need for division of labour and cooperation as well, among the cells composing their bodies. Although cells maintain their individuality for more basic biological processes, their integrated and cooperative activities improve their performance. This became one of the important principles in the establishment of different grades of body organisation. Groups of cells similar in structure and function became organised into tissues, several different tissues organised into organs to perform as the structural and functional units of body, and groups of organs, in turn, started to function together as organ systems to carry out the major activities of the body. In Chapter 18, you have come across with different kinds of organs and organ systems in earthworm, cockroach, frog and rat. The present chapter is aimed at introducing you with various types of tissues that are encountered in the multicellular animals, in general.

19.1 TYPES OF TISSUES

A **tissue** can be defined as a set of physically linked cells with similar origin along with their associated intercellular substances. They are specialised to perform a particular function or functions. However, such specialisation leading to increased efficiency of the organism requires the coordinated and integrated activities of different tissues. Here, you may recall that the branch of biology that deals with the study of tissues is called **histology** (*Gr. Histo* : web or tissue, *logos* : study).

Animal tissues can be divided into four major types, on the basis of their structure and functions. These are: epithelial tissue, connective tissue, muscular tissue and nervous tissue. All the tissues of animal body originate in the embryonic stages from any of the three primary germ layers, namely, ectoderm, mesoderm and endoderm.

19.2 EPITHELIAL TISSUES

The epithelial (Gr. *Epi :* upon, *thele :* nipple) tissue covers the external surface of the body and internal free surfaces of many organs. In an epithelial tissue, the cells are arranged very close to each other with very little extracellular material. Neighbouring cells are held together by cell junctions. The epithelial cells rest on a noncellular basement membrane, which separates the epithelium from the underlying connective tissue. Besides, there is no blood vessel supplying nutrition to the epithelial cells. They receive nutrients from the underlying connective tissue. The lining of the skin, hollow organs like the alimentary canal and the blood vessels, digestive glands like the pancreas and the liver, respiratory system, etc. are covered by epithelial tissue. The principle functions of epithelial tissues are the covering and lining of surfaces (e.g., skin), absorption (e.g., the intestine), secretion (e.g., the epithelial cells of glands), sensation (e.g., neuroepithelium) and contractility (e.g., myoepithelium). Based on the structure and function, epithelial tissues are divided into two main groups: **covering epithelium** and **glandular epithelium**.

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Covering Epithelia

These include those epithelial tissues in which the cells are arranged in layers to cover the external surface or lining of the cavities (free surface) of the body. Covering epithelia are classified into **simple** and **compound** epithelia (Fig.19.1).

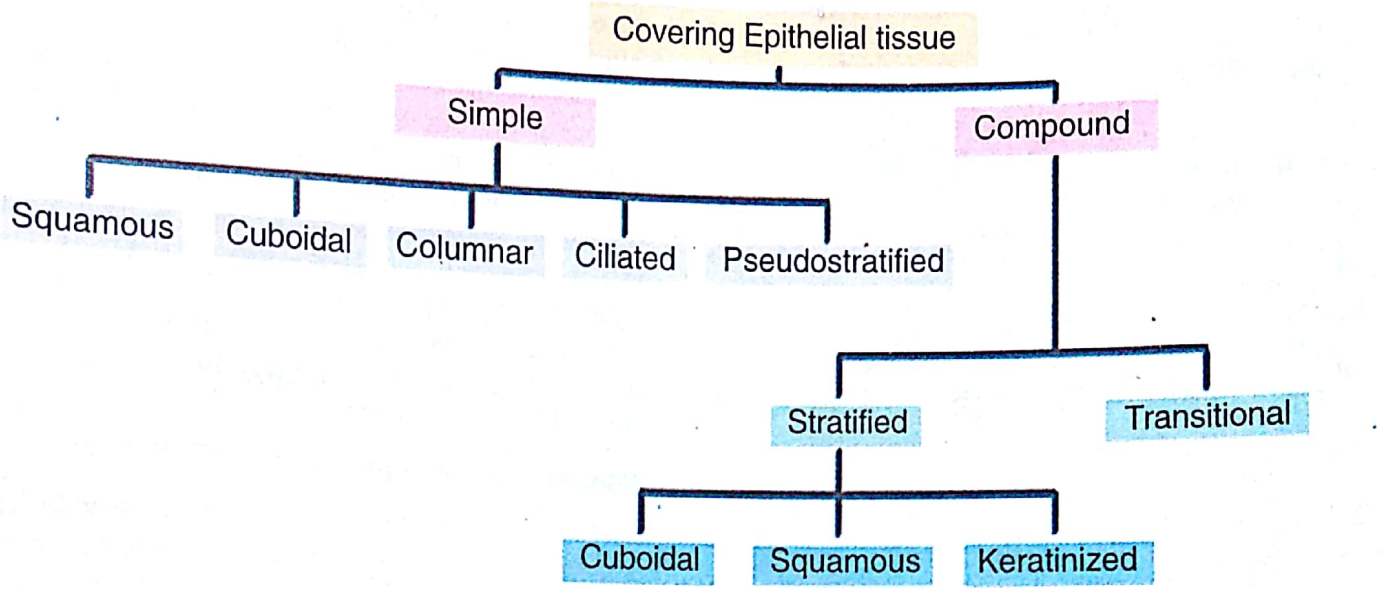

Fig. 19.1 *Classification of epithelial tissues*

Simple Epithelium

It is found generally on secretory and absorptive surfaces. It seldom covers surfaces exposed to mechanical or chemical abrasions because it is not effective in protecting the underlying tissues. It is formed of a single layer of cells resting on the basement membrane. There are various types of simple epithelia. These are: squamous, cuboidal, columnar, ciliated and pseudostratified.

Squamous epithelium consists of cells, which are thin, flat and polygonal and with prominent round or oval nucleus (Fig.19.2a). The cells have irregular boundaries that fit closely into those of neighbouring cells. For this reason, such an epithelium is also called as pavement epithelium. Its main function is protection of the underlying tissue. It forms the inner lining of lung alveoli, blood vessels, and peritoneum of body cavity. The outermost layer of skin of frog is also made up of squamous epithelium.

Cuboidal epithelium is made up of cells, which are polygonal in outline but appear cubical or cuboidal in vertical section (Fig.19.2b). In addition to protection, these cells participate in secretion (gastric juice, hormones, etc.), excretion and absorption. The cells of cuboidal epithelium in absorptive surfaces often bear microvilli on their free ends. This type of epithelium is found in proximal tubules of kidneys, lines of small salivary glands, pancreatic ducts, thyroid follicles and ovary.

Columnar epithelium is characterised by the presence of tall pillar-like cells, which resemble polygonal columns. The oval nucleus is generally present at the base of the cell. The function of columnar epithelium is secretion or absorption and it is found in the inner surface of the intestine, stomach and gall bladder. It also occurs in gastric and intestinal glands. The intestinal mucosa is lined by columnar epithelium, whose free (luminar) ends are thrown into tiny finger-like projections (microvilli). This type of projections, which are formed to increase the absorptive surface, are often referred to as brush-border.

Ciliated epithelium is formed of columnar or cubical epithelial cells that bear thin protoplasmic processes called cilia on their free

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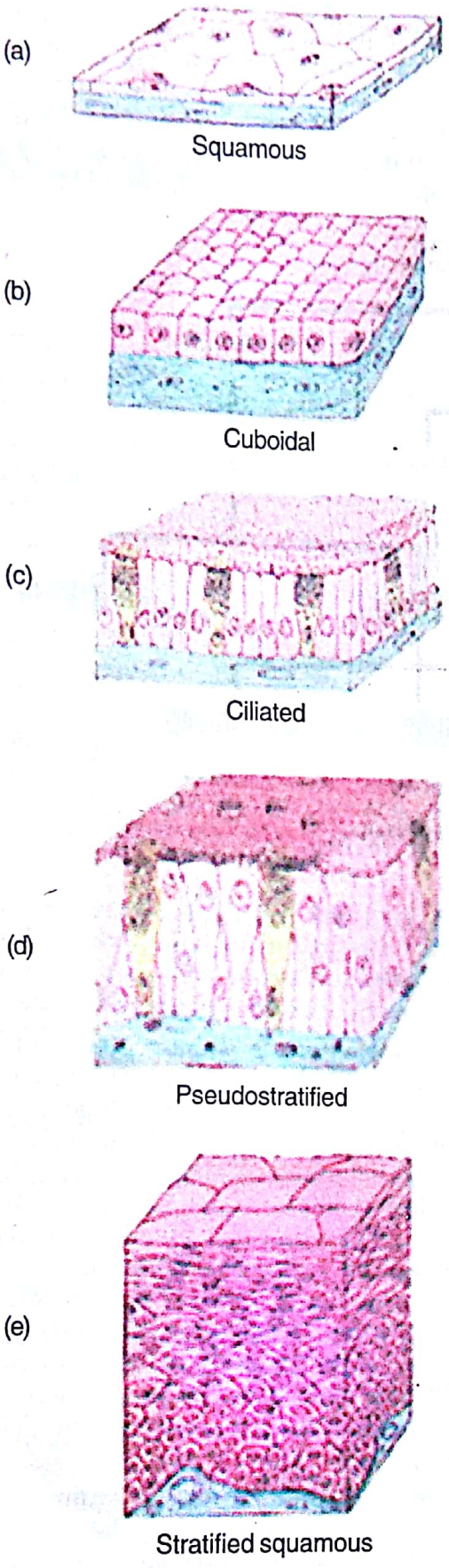

Fig. 19.2 Different types of epithelial tissues

surfaces (Fig. 19.2c). Such an epithelium is known as **ciliated epithelium**. The function of the cilia is to move particles, free cells or mucus in a specific direction (outward) over the epithelial surface. Ciliated epithelium lines the inner surfaces of some hollow organs such as fallopian tubes, nasal passage, bronchioles and small bronchi, etc.

Pseudo-stratified epithelium consists of ciliated or non-ciliated epithelium in which the cells are columnar and arranged in single layer, but appears multi-layered because some cells are shorter than others. As a result, their nuclei become located at different levels giving a stratified appearance. This type of epithelium is, therefore, called **pseudo-stratified epithelium** (Fig.19.2d). The longer cells may bear cilia. This type of epithelium covers the inner linings of trachea, large bronchi, etc. and helps to remove mucus.

Compound Epithelium

It consists of more than one layer of cells and gives a stratified appearance. Hence, these are also known as stratified epithelium. The cells may be of different shapes in different layers. The deepest layer of cells rests on a basement membrane. The morphology of the superficial layers varies in the different kinds of stratified epithelia. Compound epithelium may be stratified cuboidal, stratified squamous, stratified keratinized or transitional. Being multi-layered, compound epithelium has little role in secretion or absorption. The main function of this type of epithelium is protection to underlying tissues against mechanical, chemical, thermal or osmotic stresses. Many kinds of variation are found in compound epithelium.

In **Stratified cuboidal epithelium,** the superficial cells are cuboidal. It lines the inner surfaces of larger salivary and pancreatic ducts. **Stratified squamous non-keratinized epithelium** (Fig.19.2e) has several layers of interlinked cuboidal or columnar cells. Its cells lying closer to the underlying connective tissue are cuboidal or columnar but the superficial cells are flattened, thin walled and squamous; they retain their nuclei. It covers moist surfaces such as buccal cavity, pharynx, oesophagus, cornea of eye, etc. When the surface cells are dead and contain insoluble protein (keratin) deposits, the

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tissue is called **stratified squamous keratinized (cornified) epithelium**. This type of epithelium protects the epidermis of skin, hair, horn, nail, etc.

Another special type of compound epithelium is the **transitional epithelium**, which lines the inner surface of the urinary bladder, ureter, etc. Transitional epithelium is much thinner and more stretchable than the stratified epithelium. It has a single layer of cuboidal cells at the base; two to three middle layers of a large polygonal or pear-shaped cells and a superficial layer of large, broad, rectangular or oval cells. The cells are seen in the transitional phase, that is, they can change from simple epithelium to compound epithelium and, hence, the name is so. It prevents loss of water by reabsorption and allows considerable expansion of the organs by flattening and stretching to accommodate urine.

Histologists have identified two more types of epithelia, which cannot be included in any of the types of covering epithelia. One is the **neuroepithelial cell** of epithelial origin. These cells are specialised for sensory functions (e.g. cells of taste bud). The other one is **myoepithelial cell,** which are branched cells that contain muscle proteins, such as myosin and actin. This group of cells is specialised for contraction of the muscle cells of sweat glands, mammary glands and salivary glands.

Glandular Epithelia

These are specialised epithelial cells that form glands. Glands produce a fluid secretion that is different from blood or any extracellular fluid in its composition. Such secretions occur along with the synthesis of intracellular macromolecules, such as proteins (in pancreas), lipids (in adrenal glands), complexes of carbohydrates and proteins (in salivary glands) or all the three types of macromolecules (in mammary glands).

The cells of glandular epithelia are generally columnar or cuboidal. The glandular epithelium can be classified into two types: **unicellular**, consisting of isolated glandular cells (e.g. goblet cell of alimentary canal), and **multicellular**, consisting of cluster of cells. A gland, with a single unbranched duct is called a **simple gland** (Fig.19.3a). The secretory part of the gland consists of epithelial cells arranged in the form of tubes (tubules) or sacs (acini, alveoli) or a combination of both. The duct is also made up of

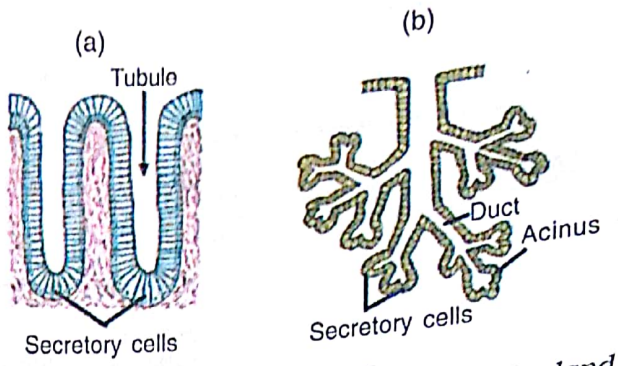

Fig.19.3 *Types of exocrine glands. (a) Simple gland and (b) compound gland*

epithelial cells. Tubular gland, found in the human intestine, is an example of simple gland. A gland with a branched system of ducts is called a **compound gland** (Fig. 19.3b). In these glands, the secretory tubule or acinus may be coiled or branched and opens into the single duct of the gland. Compound glands are present in the pancreas and sub-mandibular salivary glands.

On the basis of mode of pouring of their secretions, glands may be divided into two categories namely, exocrine and endocrine. **Exocrine glands** have a secretory portion, which contains the cells for secretion, and **ducts**, which transport their secretions to the respective sites of action, for example, salivary gland, tear gland, gastric gland and intestinal glands. **Endocrine glands** do not retain their ducts and their secretions (hormones) are picked up and transported to their site of action by the bloodstream rather than by a duct; they are called ductless glands, for example, pituitary gland, thyroid gland, parathyroid and adrenal glands. When a gland performs both exocrine and endocrine functions, it is called a **mixed gland** (e.g. the pancreas). In **holocrine glands** (e.g. sebaceous gland), the product of secretion is shed with the whole cell leading to its destruction. When the secretory granules leave the cell by exocytosis with no loss of other cellular material, the glands are called **merocrine glands** (e.g. the pancreas). In **apocrine glands** (e.g. mammary gland), only the apical portion of the cytoplasm is discharged along with the secretory product.

19.3 CONNECTIVE TISSUE

The name connective tissue is derived from its property to connect and bind different tissues or organs. It also provides the structural framework

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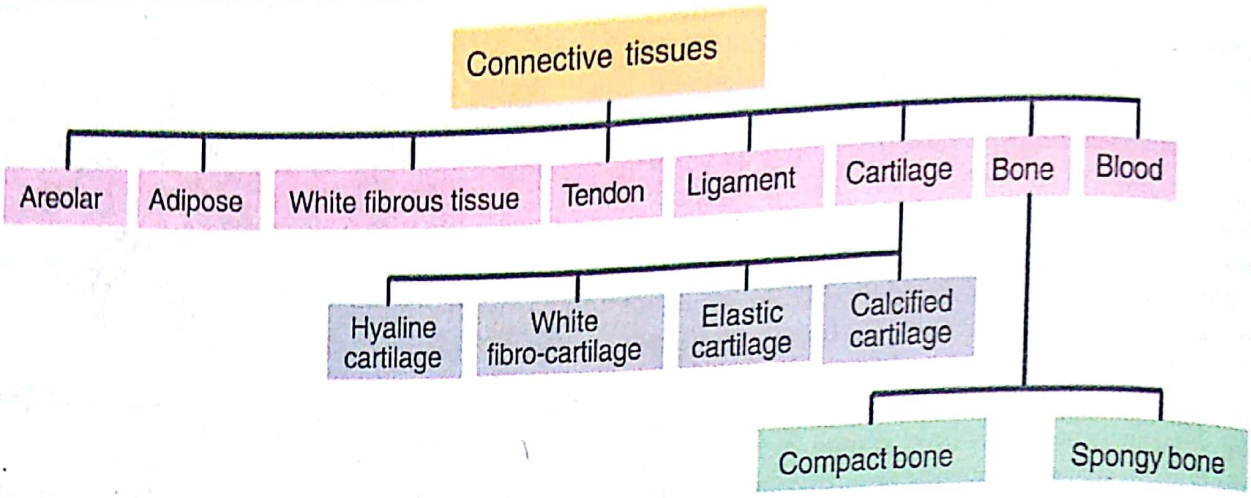

Fig. 19.4 *Classification of connective tissues*

and mechanical support for providing form of the body. It also plays a role in body defense, tissue repair, fat storage, etc. It is composed of two basic components, namely, cells and their surrounding extracellular matrix. The cells are loosely arranged and lie embedded in the matrix, which consists of protein fibres, amorphous ground substance and tissue fluid. The ground substance is formed mainly of two classes of components: **glycoaminoglycans** and structural **glycoproteins**.

There are eight types of connective tissues found in animals namely areolar, adipose, white fibrous tissue, tendon, ligament, cartilage, bone and blood. A broad classification of connective tissues is given in Figure 19.4.

Areolar Tissue

Areolar tissue, also called loose connective tissue, is found beneath the epithelia of many visceral organs, for example, stomach, trachea, skin, etc. and on the walls of arteries and veins. The areolar tissue joins different tissues, forms the packing between them and helps to keep the organs in place and in normal shape. Three types of cells are present in the areolar tissue – **fibroblasts, macrophages** and **mast cells**. Fibroblasts are the main cells of this tissue. They are irregularly shaped flat cells with long protoplasmic processes (Fig.19.5). Fibroblasts synthesise two kinds of proteins – **collagen** and **elastin**. The fibres of areolar tissue are of two types, namely, **white collagen fibres** and **yellow elastin fibres**. The collagen fibres are made up of collagen proteins and are found in the form of wavy bundles lacking elasticity. Yellow elastin fibres are elastic, translucent, made up of elastin protein, and are not present in bundles. These form a kind of lattice by joining their branches. The tensile strength of collagen fibres and the elasticity of elastic fibres prevent displacement and injury of tissues and organs in which areolar tissues are present. During tissue repair and wound healing processes, collagen fibres are formed at the sites of injury. Macrophages (large amoeboid cells) are another component of the areolar tissue, which destroy microbes, foreign particles, and the cells of damaged tissues by phagocytosis. The third cellular component of areolar tissue is made up of mast cells, which are large, irregular and ovoid cells. These store inflammation-producing substances, which are released in the vicinity of injury to attract phagocytes. This is the reason why inflammation takes place near the site of injury.

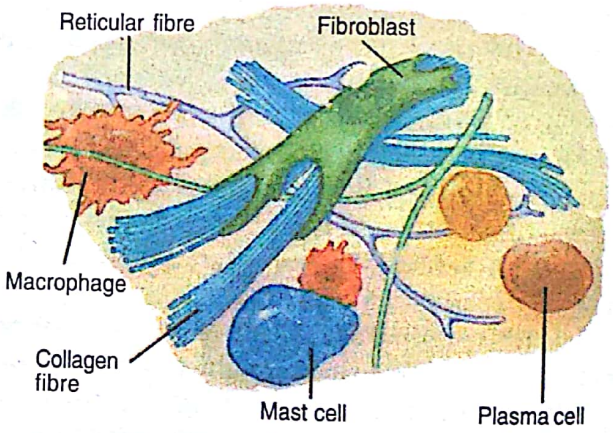

Fig. 19.5 *Areolar tissue*

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Adipose Tissue

Adipose tissue is a connective tissue present beneath the skin, around kidneys and in mesentery and bone marrow. It is characterised by the presence of **adipocytes** as cellular component, which are large, spherical, or oval cells also called fat cells. The cytoplasm and organelles in adipocytes are pressed by fat into a narrow annular layer just beneath the plasma membrane. Besides adipocytes, it contains **fibroblasts, macrophages, collagen fibres** and **elastic fibres** (Fig. 19.6). The adipose tissue synthesises, stores and metabolises fat. It is a very prominent component of skin in mammals living in polar regions. It prevents heat loss by forming a heat-insulating layer beneath the skin. It forms shock-absorbing cushions around kidneys and eyeballs.

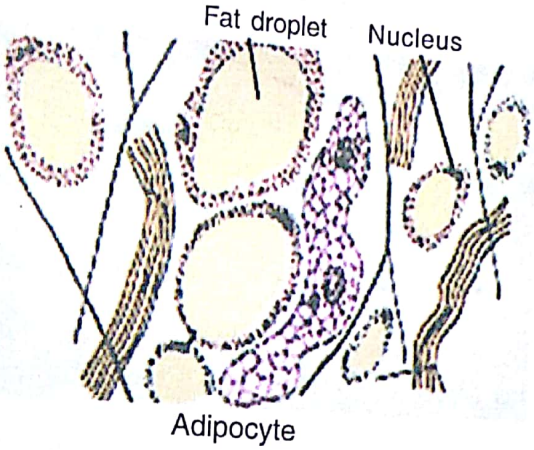

Fig.19.6 *Adipose tissue*

White Fibrous Tissue

This specialised connective tissue is provided with only a few **fibroblasts** (cellular component) that are scattered amidst the dense network of thick collagen fibre bundles (matrix). It has great tensile strength. The presence of white fibrous tissue at the joints between skull bones makes them immovable.

Tendon

It is a dense, strong fibrous connective tissue with thick, parallel, bundles of **collagen fibres** (matrix). A few flat and elongated fibroblast cells lie in single rows between the fibre bundles. Tendon forms the strong inextensible attachment of a skeletal muscle to a bone (Fig.19.7).

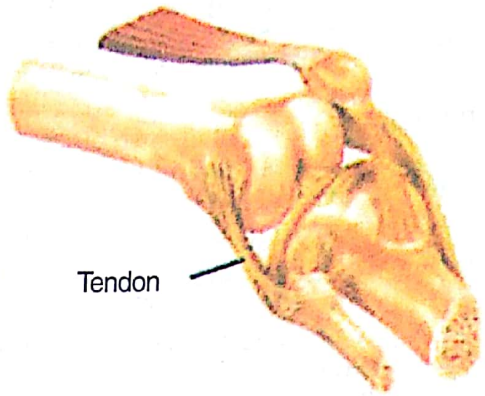

Fig.19.7 *Section through the knee joint showing tendon*

Ligament

It is a dense fibrous connective tissue. Its ground substance or matrix is densely crowded with **yellow elastin fibres** branched in different directions. A few elongated flat cells (fibroblasts) lie scattered between the fibres. The ligament connects bones at the joints and holds them in position. Because of the fact that ligaments connect the bones at joints, we are able to move and rotate our neck, limbs, fingers, etc. comfortably.

Cartilage

Cartilage is a solid but semi-rigid and flexible connective tissue. Its matrix is rich in **chondrin,** which consists of **proteoglycans** (protein chain bonded to long chains of disaccharide hyaluronic acid units). **Chondrocytes** are large and bluntly angular cartilage cells lying scattered in the matrix. They occur in clusters of two or three cells in small spaces called lacunae (Fig. 19.8).

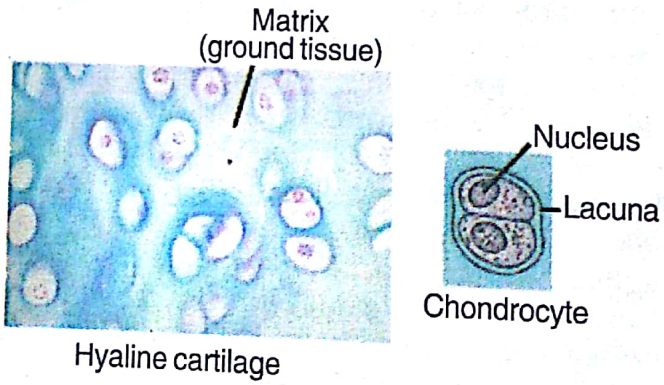

Fig.19.8 *Hyaline cartilage*

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Four types of cartilage are found in animals. These are **hyaline**, **fibrous**, **elastic** and **calcified**. (i) Hyaline cartilage- its matrix is clear, homogeneous, translucent and bluish in colour, lacking fibres. It is present in sternum, hyoid and ribs, etc. (ii) Fibrous- it contains matrix rich in bundles of collagen fibres along with chondrocytes. This can be seen in intervertebral disc. (iii) Elastic- this is similar to fibrous cartilage except for yellow elastin fibres in place of white collagen fibres. This type of cartilage is elastic and can be seen in pinna, tip of nose and epiglottis. (iv) Calcified- initially it is like hyaline cartilage but later on it gets hardened like bone due to deposition of calcium salts, for example, supra scapula of frog's pectoral girdle, pubis of pelvic girdle of frog, etc.

Bone

Bone is a solid, rigid and strong connective tissue. The matrix is composed of a protein called **ossein.** Matrix of bone is rich in phosphate, sulphate, carbonate, and fluoride salts of calcium and magnesium. Flat irregular spaces called lacunae occur in the solid matrix. Each lacuna contains a flat bone cell or **osteocyte.** An osteocyte has an irregular shape and long cytoplasmic processes. These processes extend into minute canals (canaliculi) radiating from each lacuna. Compact bone forms the dense outer layers of all bones. It is composed of many parallelly and longitudinally arranged column-like structures called **Haversian system**, which remain cemented to each other (Fig. 19.9). In each Haversian system, several concentric layers (Lamellae) of bony matrix encircle a longitudinal central canal (Haversian canal). This canal carries blood vessels and nerves. Lacunae containing osteocytes occur in a layer between two lamellae. The hollow cavities or spaces are called marrow cavities. Marrow cavity filled with adipose tissue is called **bone marrow**. Bones are of two types, namely compact and spongy. **Compact bones** represent the shaft of long bones and have lamellae arranged in Haversian system without gaps. **Spongy bones** are found in vertebrae, ribs, skull and epiphysis of long bones. In spongy bones, lamellae called trabeculae are present. These form interlacing network with small spaces between them. These spaces contain **red bone marrow** that is highly vascularised and forms erythrocytes and granular leucocytes. Red bone marrows are found in ribs, vertebrae and skull bones. The yellow bone marrow is composed of fatty tissue. It stores fat and produces blood corpuscles only in emergency situations. It is found in the central part of long bones.

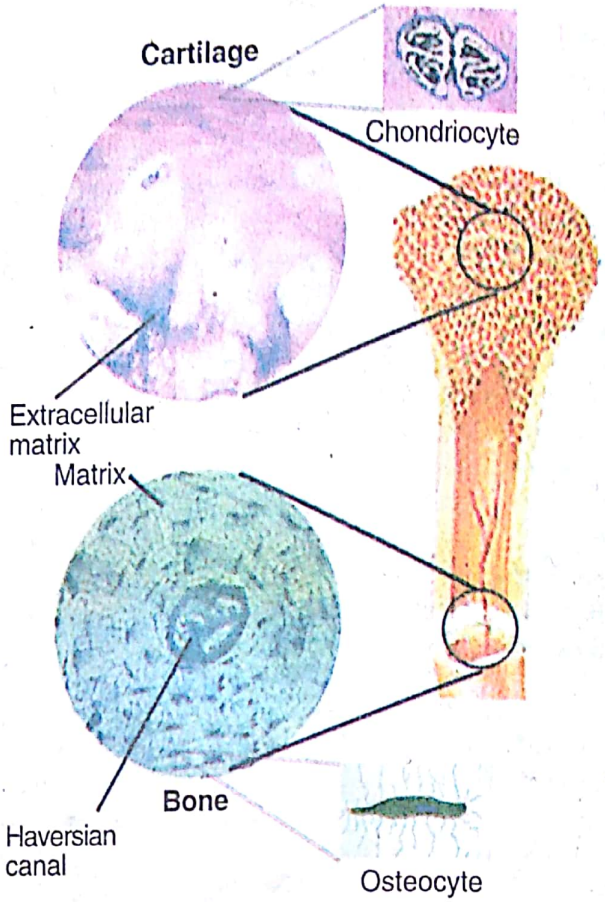

Fig.19.9 Internal structure of a bone

Blood

Blood is a fluid **connective tissue.** Its cells are quite distinct from other connective tissue cells, both in structure and function. There are different types of cells with very specific functions and are free floating in a fluid called **plasma**. Plasma is equivalent to matrix of other connective tissues but lacks fibres. The matrix of other connective tissues is stationary whereas matrix of blood is non-stationary and free flowing. That is why blood is also called liquid tissue. It is also called vascular tissue because it is part of the vascular system of many animals.

Plasma is faint yellow in colour and slightly alkaline (pH 7.4). It is somewhat viscous and an aqueous solution containing many organic